COLORADO RIVER BENTHIC FOODBASE STUDIES IN GLEN AND GRAND CANYONS: YEAR 1 FINAL REPORT



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Museum of Northern Arizona

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EXECUTIVE SUMMARY

INTRODUCTION

The Colorado River ecosystem (CRE) between Glen Canyon Dam and Lake Mead has been highly altered by Glen Canyon Dam (Hofnecht 1981, Blinn and Cole 1991, Angradi 1994, Stevens et al. 1997, Schmidt et al. 1998, Cross et al. 2013, Kennedy et al. 2016). Largely missing from the river and of particular concern are the "EPT" taxa, including mayflies (Ephemeroptera), stoneflies (Plecoptera), and caddisflies (Trichoptera), and other benthic macroinvertebrate (BMI) taxa that are important elements of fisheries foodbases and often are used as biological indicators of stream ecosystem health (Barbour et al. 1999, Merritt et al. 2008). In addition, the benthic sediments of Lake Powell Reservoir and the Glen Canyon Dam CRE tailwaters are conspicuously anoxic from the dam downstream to the Paria River confluence. We conducted research to address two objectives in 2016-2017 to: 1) identify the suite of EPT species that could occur in the Glen Canyon Dam tailwaters, and integrate understanding of their life histories and water quality requirements; and 2) evaluate the distribution, causal factors, and impacts of benthic and hyporheic anoxia (BHA) on macroinvertebrate habitat and colonization potential. We present the results of these studies as a contribution toward understanding the ecological conditions and potential management options for the Glen Canyon Dam tailwaters and downstream CRE in Grand Canyon.

METHODS

Objective 1 – Potential EPT species in the Glen Canyon Dam tailwaters and CRE

We compiled EPT distribution information for species potentially occurring in the Glen Canyon Dam tailwaters and the CRE and the larger Grand Canyon ecoregion (GCE) from multiple sources, including the literature, by data-mining regional and national museum collections, and analyses of state-based range distribution data (Appendix 1A). We compiled environmental tolerance values for each EPT genus from Barbour et al. (1999) and Merritt et al. (2008; Appendix 1B). Biogeographic affinity and occurrence data also were compiled from state-based range data as well as literature sources.

Objective 2 – Investigate BHA distribution and seasonal change in the Colorado River in the Glen Canyon Dam tailwaters

The study area for field investigations of BHA included lower Lake Powell (the forebay of the dam) and the Glen Canyon Dam tailwaters from the base of the dam to approximately two kilometers below Lees Ferry (including the mouth of the Paria River). Five tasks were accomplished in this component of the study. 1) The distribution of BHA was measured at 12 transects, including three sites in lower Lake Powell reservoir, six sites in the Glen Canyon reach from Glen Canyon Dam downstream to Lees Ferry, and three sites downstream from Lees Ferry to CR 1R. 2) A leveling instrument and survey elevation rod were used seasonally to document and monitor the presence and distribution of BHA, and measure selected water and sediment geochemical variables on the transects. 3) The effects of BHA water and sediment geochemistry were used in field and laboratory experiments of different treatment and temporal combinations to test the survivorship of several common benthic macroinvertebrate taxa from

in and around the study area in relation to BHA exposure. 4) Field experimental data and analysis of the 2016 high flow experiment from Glen Canyon Dam were used to evaluate the impacts of mechanical disturbance on BHA development rate and BMI responses. 5) Lastly, we experimentally investigated the factors influencing BHA development using fluctuating versus nonfluctuating flow field mesocosms, and conducted a three-treatment (temperature, aeration, and macrophyte presence) laboratory experiment to test the rate of BHA development under controlled conditions.

RESULTS AND DISCUSSION Objective 1: Potential EPT in the CRE

The list of actual and potentially occurring EPT species in the Grand Canyon ecoregion, the drainage basin of Grand Canyon, includes 382 taxa, including 104 Ephemeroptera taxa among 41 genera in 10 families; 95 Plecoptera species among 44 genera in 8 families; and 185 Trichoptera species among 64 genera in 21 families (Appendix 1A). In Grand Canyon, we report a possible or detected total of 18 Ephemeroptera species among 15 genera in 6 families; 6 Plecoptera species among 5 genera in 3 families; and 45 Trichoptera species among 25 genera in 15 families. Thus, the EPT fauna in the overall GCE is relatively robust, including the tributaries in Grand Canyon with the exception of Plecoptera. While the diversity of EPT in tributary streams is relatively high, it remains depauperate in the Colorado River mainstream. *Rhyacophila coloradensis*, one or more hydroptilids, and several other species are the only caddisfly species regularly encountered in the CRE, and may be useful as indicator species.

Analyses of water quality requirements, habitat, and feeding strategies (where known) indicate that many of the EPT species reported or potentially occurring in the CRE and GCE appear to be capable of existing in some portion of the CRE mainstream under conditions of low flow variability and high water transparency (Appendix 1B). However, all Plecoptera except 3 species of Perlidae (*Hesperoperla* and *Isoperla*) occur at elevations far above that of the mainstream, and at least 101 GCE Trichoptera species (55%) occur at elevations above 1200 m and therefore at higher elevations than the mainstream. Although water temperatures are generally cooler at higher elevations, other water quality variables, such as conductance, often differ substantially from those in the CRE (e.g., Ledbetter et al. 2016). In addition, CRE suspended sediment loads, thermal constancy, benthic habitat structure, and daily flow fluctuations also may limit EPT colonization (Cross et al. 2013, Kennedy et al. 2016). Understanding the influences of those and other factors on individual EPT species life histories and potential for colonization success in the Glen Canyon tailwaters reach will require detailed investigations. Our data indicate that factors other than water quality limit EPT colonization and survival in the mainstream Colorado River downstream from Glen Canyon Dam.

Objective 2: BHA Distribution, Development, and Ecosystem Impacts

2a, b - **Distribution and Seasonal Shifts:** Collectively, the field and laboratory measurements and experiments clarified the distribution, developmental rate, and general impacts of BHA on the Colorado River ecosystem. BHA developed abundantly in fine sediments below the low water stage in the reservoir and in the river upstream from the Paria River confluence, particularly in low velocity settings dominated by the presence of extensive benthic

macrophyte cover (*Chara* nr *vulgaris*, *Zanichellia* sp., other macrophytes). Little BHA was detected in the thalweg of the mainstream channel, suggesting that photosynthetically active radiation (PAR) limitation with higher velocities and reduced macrophyte cover limited BHA development. Limited but detectable BHA development occurred beneath gravels and cobbles in cobble bar habitats (e.g., CR -9R) upstream from the Paria River confluence, but not downstream in Grand Canyon.

BHA distribution varied seasonally in the reservoir and to a lesser extent in the tailwaters, developing relatively gradually following prolonged summertime inundation, and occurring at inundated higher stage elevations during the late summer and autumn months. BHA dissipated when reservoir and downstream river shoreline stages decreased in elevation for more than 12 hr during seasonal dewatering (e.g., late autumn). Although seasonal shoreline shifts in BHA were erratic, perhaps due to high flow experiment (HFE) impacts, BHA advanced shoreward at Lees Ferry by 1.49 m from summer to fall, 2016, and then retreated with low late winter and springtime flows. The development of BHA at Site CR-3 likely was due to the dense wetland vegetation and nearly constant inundation in the shallow return current channel depression there. Overall, the extent of BHA development along shorelines was greatest during early autumn, occurring further upslope during fall, and dissipating during winter and springtime.

BHA was not detected at the three sites below Lees Ferry during the summer sampling period. However, traces of BHA were found among the cobbles at CRBLF1 and in shoreline sand deposits at CRBLF2. The furthest downstream site, CRBLF3, developed considerable amounts of BHA to >30 cm depth following Paria River flooding and deposition of extensive quantities of organic matter. However, those sediments and organic deposits were flushed from that eddy during the 2016 HFE, and BHA did not re-develop during the 2017 winter and springtime periods when benthic macrophyte production was low. Farther downstream, BHA was rarely detected in a few low-velocity tributary mud deposits during several river trips in 2016 and 2017. The duration and extent of BHA development in the CRE in Grand Canyon and its perennial tributaries is limited, likely due to the absence of extensive aquatic macrophyte cover, high dissolved oxygen concentrations, and dynamic sediment transport and flow regimes.

Water Column and BHA Sediment Geochemistry: Laboratory water quality analyses indicated little detectable mercury (Hg) or uranium (U) mobilization in BHA-dominated sediments, with nearly all samples having below minimum detectable levels, except for sediments from Lees Ferry and CR-12. At Lees Ferry, contrary to our expectations, the Hg concentration was 5 μ g/L in BHA sediments, but was higher (7 μ g/L) in non-BHA sediments. The EPA has established a maximum contamination limit (MCL) for Hg in drinking water of 2 μ g/L (https://www.epa.gov/wqc), while the World Health Organization limit is 6 μ g/L. For acute (maximum) and chronic (continuous) freshwater aquatic systems, the EPA has established a limit of 1.4 and 0.77 μ g Hg/L, respectively. Our field sediment values at Lees Ferry were 2.5- to 3.5-fold higher than EPA MCL for drinking water, and were slightly higher than the World Health Organization's drinking water MCL. Those sediments were 3.6- to 5-fold higher than acute levels, and 6.5- to 9.1-fold higher than chronic exposure in fresh waters.

Two of the three water samples (LP1 and CRBLF3) analyzed for U revealed U concentrations at or below reported detection limit (0.01 mg/L). However, the CR-12 sample

contained 0.019 mg/L U, slightly above the reported detection limit but below the EPA's maximum contamination limit (MCL) for drinking water (MCL = 0.03 mg/L).

Water quality analyses revealed no detectable release of nitrate, nitrite, or sulfate from BHA sediments. However, during the summer sampling period, we detected increased total P concentration in water extracted from BHA sediments. BHA sediment samples similarly revealed slightly elevated total P, and also increased nitrate, total C, and organic matter compared to non-BHA sediments at CR-6.5 and LP1. However, the opposite pattern was documented at CRLF, where BHA sediments contained lower concentrations of total P, nitrate, total C, and organic matter compared to non-BHA sediment samples.

BHA impacts on Colorado River water quality appear to be slow and trivial at a microsite scale, based on laboratory analyses of field water column and BHA sediment sampling. Our results are consonant with the findings of Wildman et al. (2010) in Lake Powell Reservoir, who reported minor seasonal releases of U from benthic sediments. However, low levels of geochemical activity within BHA sediments or at the sediment-water interface may result in cumulative, long-term, element-specific release or sequestration. In addition, toxic compounds may be released from mobilized BHA sediments during HFEs, a phenomenon that may warrant further study.

2c - Benthic Macroinvertebrate Responses: Bioassay experiments were conducted to determine the effects of water from BHA sediments on existing and potential benthic macroinvertebrate species. We subjected Heptagenia mayfly larvae to 10-day treatments of BHA sediment with Colorado River water against controls without BHA sediments. This experiment generated significant negative effects on *Heptagenia* survival in the BHA treatment $(t_{0.05, 19} = 2.213, p = 0.039)$. By the fifth day, all mayflies in the BHA treatment had died, whereas only 40% of mayflies in the control treatment had perished. Water quality differed as well, with electrical conductivity and DO differing between the two treatments. The 50 percent mortality rate (LD₅₀) in the BHA treatment occurred at 18.3 hrs after the start of the experiment, and LD₅₀ in the control group was expected to occur at 562 hrs after the start of the experiment (the experiment was discontinued after 10 days). Heptagenia mayfly larvae are flowing water species, and are more sensitive to poor water quality than are damselflies or amphipods. No mayfly species are presently reported in the mainstream of the Colorado River in Glen or Grand Canyons, but are found in isolated side pools. The high degree of mortality of one mayfly species caused by BHA sediments may be a factor contributing to their absence in the mainstream, particularly in low velocity settings where the fine BHA particles may physically obstruct respiration in mayfly gills, leading to mortality. Thus, this physical impact may more strongly limit mayfly presence than do the chemical properties of BHA sediments.

2d – Flow and Flooding Impacts: Bed disturbance was experimentally simulated by raking three 9 m² patches in lentic and cobble bar habitats in August 2016. The patches were initially black in color, but within 12 hr had undergone oxic transition. The lentic (sand-floored) channel setting at CR -6.5R developed a 1 cm buff sand surface layer that persisted until the onset of the November 2016 HFE (below). The CR -9R cobble floored channel similarly lost its black anoxic appearance, and remained visible for more than a month. Experimental caging of benthic macroinvertebrates onto treated and untreated benthic surfaces did not reveal clear

differences in mortality, probably because the organisms used (*Gammarus*, *Physa* and New Zealand mud snails) are highly tolerant of BHA conditions.

The post-HFE transect surveys at CR-6.5 and CRLF indicated a minor recession of BHA distribution in the near-shore habitat as a result of artificial flooding. Prior to the HFE, BHA was found 0.18 m (at LF) and 0.61 m (at CR-6.5) closer to shore than during the post-HFE period, indicating a small amount of scour and aeration of the sediment resulted from the HFE. This minor recession suggests that HFEs can reduce BHA to some extent. In addition, the HFE scoured some *Chara* cover and some of the decomposing macrophytic organic matter from the bed, resulting in development of a 1 cm-thick buffer layer of oxic sand throughout the tailwaters. This buffer layer persisted into the May transect survey in 2017.

Collectively, these results indicate that fine-scale or minor system-wide artificial disturbances can regenerate surficial oxic conditions on the bed that may last for biologically meaningful time periods; however, the subsurface sediments beneath the buffer layer retained their blackened BHA character, and without further disturbance, BHA gradually becomes re-established. Increased frequency, duration and flow volume may have longer-lasting effects on reducing BHA below Glen Canyon Dam, but further research into the specifics of HFEs that would be most beneficial is warranted.

2e - **Development of BHA:** A combination of field and laboratory experiments were conducted to determine the rate and factors responsible for development of BHA. A field mesocosm experiment revealed significant development of BHA within a 38-day experimental period. BHA developed only in chambers into which *Chara* had been added. BHA development in that experiment occurred to an equal extent in chambers with *Chara* under both steady (floating) and unsteady (shoreline) flows; experimental chambers containing clean sand without Chara did not develop visibly detectable BHA. Water quality in experimental chambers with *Chara* had decreased dissolved oxygen concentration, pH, and oxidation-reduction potential (ORP) in both treatments, indicating that *Chara* influenced those water quality variables, leading to the development of BHA. This initial field experiment emphasized the importance of macrophytic vegetation on BHA development, and suggested that the minor flow fluctuations (0.5 m/day) and wave action do not greatly influence BHA developmental.

We then conducted a three-treatment laboratory experiment to distinguish the individual and interactive contributions of water temperature, aeration, and the presence of *Chara* on BHA development. All three treatment variables significantly affected the development of BHA (measured as depth of darkened sediment at the end of the experiment): *Chara:* $F_{1,84} = 1244.75$, p < 0.001; temperature: $F_{2,84} = 12.77$, p < 0.001; aeration: $F_{1,84} = 21.96$, p < 0.001. No BHA development occurred in the absence of *Chara*, and sediment in all chambers with *Chara* present developed conspicuous BHA. This indicates that large quantities of decaying *Chara* (and possibly other types of aquatic vegetation) is necessary for anoxia development. In addition to the presence of *Chara*, higher temperatures ($T_{20} > T_5$, p < 0.001; $T_{20} > T_{12}$, p = 0.037; $T_{12} > T_5$, p = 0.0341) and lack of aeration (non-aerated > aerated, p < 0.005) led to increased BHA

In addition to effects of each variable independently, significant interactions occurred among all variables with respect to BHA development. Therefore, in summary, the combination of warm water temperature, the presence of *Chara*, and the lack of aeration promoted most rapid development of BHA in benthic sediments. Altering any of these three factors (i.e., decreasing water temperature, removing *Chara* biomass, or increasing aeration), or tandem combinations thereof, are likely to retard, but not prevent or fully reverse BHA development.

CONCLUSIONS AND MANAGEMENT OPTIONS

This preliminary study provides evidence for the mechanistic underpinnings of the development and maintenance of BHA in the Colorado River in Glen Canyon, and some potential ecosystem consequences of the phenomenon. Several separate but interrelated management actions may be used to decrease the impacts of BHA in Glen Canyon. These include: 1) increasing mainstream DO concentration, 2) increasing mainstream turbidity, 3) decreasing mainstream temperature, and 4) increasing mainstream flow variability. Some of the many strategic issues and tradeoffs associated with these management options are discussed, and more consideration and study is needed prior to taking management action.

Many uncertainties remain after this pilot examination of BHA in the Glen Canyon reach. The extent of BHA development documented in the Glen Canyon Dam tailwaters is considerable, with virtually all of the river bed downslope of the 200 m³/s stage (approximately 2 km² of the channel) affected by BHA. Currently, the phenomenon appears to be limited to Lake Powell sand deposits and the tailwaters reach. BHA development is driven by temperature, flow stability, and benthic plant growth, but does not greatly alter water column geochemistry. BHA appears to be detrimental to sensitive larval macroinvertebrates like some mayfly species, possibly though physical impacts on respiration. Consequently BHA may limit the potential aquatic macroinvertebrate assemblage and foodbase in the Glen Canyon reach. BHA is not likely to develop to any great extent downstream in Grand Canyon unless suspended sediment load decreases, creating prolonged periods of water clarity, and macrophytic vegetation extensively colonizes benthic fine sediment deposits.

INTRODUCTION

PROBLEM STATEMENT

The Colorado River ecosystem (CRE) between Glen Canyon Dam and Lake Mead has been highly altered by Glen Canyon Dam (Hofnecht 1981, Blinn and Cole 1991, Angradi 1994, Stevens et al. 1997, Schmidt et al. 1998, Cross et al. 2013, Kennedy et al. 2016; Fig. 1). Largely missing from the river and of particular concern are the "EPT" taxa, including mayflies (Ephemeroptera), stoneflies (Plecoptera), and caddisflies (Trichoptera), which are important elements of threatened fish and fisheries foodbases and often are used as biological indicators of stream ecosystem health (Minckley 1991, Barbour et al. 1999, Pomeroy et al. 2000, Merritt et al. 2008). Those insect orders are relatively abundant in many unaltered tributary streams in the CRE and in upstream regulated segments. For example, EPT density is relatively high in the coolwater tributaries in Grand Canyon, in the regulated Flaming Gorge and other Colorado River reaches upstream in Utah and Colorado (Oberlin et al. 1999, Vinson 2001, Haden et al. 2003), and also in the Davis and Parker region downstream from Hoover Dam, but EPT diversity is low in the CRE in Grand Canyon (Shannon et al. 2001, Blinn and Ruiter 2009a, Kennedy et al. 2016). Many physical and biological factors are potentially responsible for the absence of EPT in the Glen Canyon Dam tailwaters, including altered seasonal water temperature variation, daily and seasonal flow variation, and legacy effects of predam sediment loads on colonization (Stevens et al. 1997, Cross et al. 2013). Kennedy et al. (2016) posit that hydropower flow fluctuations reduce benthic macroinvertebrate (BMI) egg survival, restricting the assemblage to those species that oviposit on the water's surface. Other limiting factors, some of which are dam related, may affect egg survivorship and other life stages, including water quality, habitat availability (e.g., embeddenness), suspended sediment concentration, and the development of benthic and hyporheic anoxia (BHA).

Our research involved two related objectives: 1) identify the potential suite of EPT species that occur in the Glen Canyon Dam tailwaters, and integrate understanding of their life histories and water quality requirements; and 2) evaluate the distribution, causal factors, and impacts of BHA on macroinvertebrate habitat and colonization potential. We present the results of these studies as a contribution toward understanding the ecological conditions and potential management options for the Glen Canyon Dam tailwaters and downstream in the CRE.

OBJECTIVE 1

The EPT and other aquatic macroinvertebrates of Glen Canyon and the Grand Canyon region are known from riverine and regional studies since 1959, as well as biogeographic studies of selected taxa (e.g., Woodbury et al. 1959; Stone 1964; Stone and Queenan 1967; Stone and Rathbun 1968, 1969; Allan 1975; Polhemus and Polhemus 1976; Peckarsky et al. 1985; Ruiter 1995; Spindler 1996; Sublette et al. 1998; Oberlin et al. 1999; Call and Baumann 2002; Stevens and Polhemus 2008; Stevens et al. 2008; Blinn and Ruiter 2005, 2009a, b; Stevens and Bailowitz 2009; Appendix A). Regional- or state-based distribution of southwestern EPT are known for Ephemeroptera (McCafferty et al. (2012), some Plecoptera (Stark et al. 1986, Kondratieff and Baumann 2002), and Trichoptera (e.g., Allan 1975, Herrmann et al. 1986, Weaver 1988, Rasmussen and Morse 2016), but many species in regional and national collections remain to be databased. Woodbury et al. (1959), the only significant predam study

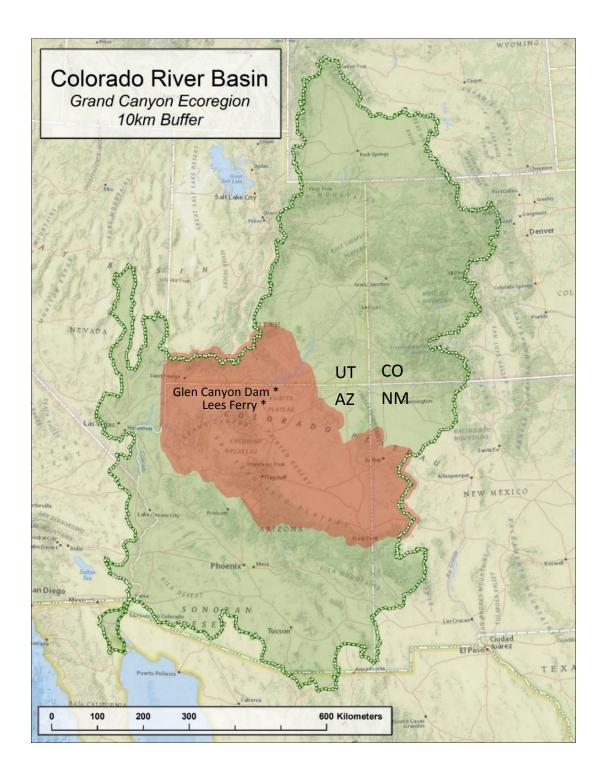


Fig. 1: Map of the Colorado River basin, indicating the upper and lower basins designated by the Colorado River Compact of 1922, and highlighting the Grand Canyon ecoregion that drains into Grand Canyon. Map by J. Jenness, Museum of Northern Arizona Springs Stewardship Institute, Flagstaff.

of the Colorado River in Glen Canyon, indicated that few macroinvertebrate species occurred in the unregulated mainstream (Stevens et al. 1997). While Woodbury et al. (1959) reported on the aquatic macroinvertebrate species collected in Glen Canyon prior to impoundment, confusion remains over which species they collected in the mainstream versus springs or tributary habitats during their survey. Here, we clarify and integrate the results of that study with taxonomic and post-dam literature on EPT distribution, as well as data derived from searches of several museums.

The specific objectives of Objective 1 were thus to compile a list of EPT species found in the Colorado Plateau region that would possibly be present in the Colorado River Basin, including the Grand Canyon Ecoregion (GCE) and the Colorado River below Glen Canyon, based on distribution and water quality requirements.

OBJECTIVE 2

Anoxia in benthic and hyporheic sediments affects aquatic benthic and hyporheic geochemistry, habitat quality, biota, and ecosystem interactions (Boulton et al. 1998; Baker et al. 2000; Clements et al. 2000), but its influence in large regulated rivers, such as the Colorado River downstream from Glen Canyon Dam, remains largely unknown. BHA often develops through excessive production and decomposition of aquatic macrophytic vegetation in habitats with high water clarity and productivity. Insufficient stream energy exists in lentic and lowvelocity settings to export decomposing organic matter, causing BHA to expand. BHA development is often conspicuous, appearing as blackened sediment or sediments, and can be detected using simple methods (e.g., Marmonier et al. 2004). However, BHA development and feedback influences on ecosystem geochemistry, macrophyte assemblages, and higher trophic level structure and function can be complex, counter-intuitive, and indirect (Dahm et al. 1987, Baker et al. 2000, Campbell et al. 2003, Fleeger et al. 2003). For example, experimental reduction of benthic sediments in a highly contaminated Australian estuary generated the expected release of Mn and Fe, but only minor releases of As, Cd, Cu, and Zn. Similarly, minor and slow release of U has been reported at the anoxic sediment-water column interface (Wildman et al. 2010). However, hyporheic anoxia in mining waste-contaminated Mill Creek, Idaho resulted in active uptake and transport of Se or selenides throughout the sediment profile (Oram et al. 2010). Release of anoxia-liberated compounds into the overlying water column can induce bioaccumulation and directly and indirectly affect aquatic assemblage, foodweb, and trophic structure and function (Cain et al. 1992, Hare 1992, Poulton et al 1995, Clements et al. 2000, Besser et al. 2001, Campbell et al. 2003, Hogsden and Harding 2012), with impacts extending into the riparian zone (Moore et al. 1991, Walters et al. 2008). Thus, BHA can influence aquatic ecosystem function, even though its influences in the highly regulated Colorado River downstream from Glen Canyon Dam have not been examined.

Minor development of BHA has been reported in the Glen Canyon Dam tailwaters (Stevens et al. 1997), and also has been studied in relation to heavy metal release in Lake Powell reservoir (WildIman et al. 2010). However, prior to the onset of highly constrained hydropeaking flows from Glen Canyon Dam in the early 1990s, BHA was not recognized as a phenomenon much influencing the regulated Colorado River aquatic ecosystem. However, since the late 1990s BHA has become a conspicuous phenomenon from Glen Canyon Dam to

the Paria River confluence downstream from Lees Ferry. This increase and distribution shift corresponds with the high levels of water clarity and benthic primary production in that reach. An abrupt reduction in mainstream water clarity and production occurs at the Paria River confluence due to fine sediment input, creating a conspicuous, stair-step reduction (30-fold) in macrophyte standing stock downstream in Grand Canyon (Graf et al. 1991, Stevens et al. 1997, Cross et al. 2013). Paria River fine sediment contributions reduce photosynthetically active radiation (PAR) in the downstream mainstream water column (Yard et al. 2005), reducing macrophyte production and the deposition of fine organic matter.

The impacts of BHA on water and benthic sediment quality and benthic macroinvertebrate survivorship in Lake Powell and Glen Canyon tailwaters have not received extensive attention prior to this study. Previous research has shown, however, that anoxia-related diffusion on Mn and U from pore water into the water column is occurring in Lake Powell (Wildman et al. 2010), and elevated concentrations of Hg and Se exist in multiple trophic levels downstream from the dam, including in fish (Walters et al. 2015). Sources and consequences of trophic contamination have not yet been determined on the river ecosystem, but the effects of metal mobilization on aquatic organisms can include: oxidative stress in algae and transport of these metals to higher trophic levels (Pinto et al. 2003); changes in benthic macroinvertebrate (BMI) assemblage composition, behavior, competition, and predation (e.g., Fleeger et al. 2003); and overall negative impacts on BMI abundance and diversity (e.g., Clements et al. 2000).

Specifically, we focused on the following tasks in Objective 2: a) seasonally map variation in the distribution of BHA in the Glen Canyon Dam tailwaters and lower Lake Powell; b) characterize water and sediment chemistry in benthic areas affected by BHA; c) determine the effects of BHA water on aquatic macroinvertebrates; d) determine effects of simulated and high flow disturbance on BHA development; and e) determine the conditions for, and developmental timing of BHA. Because this was a pilot effort to understand BHA distribution and development, we conducted an array of small-scale, exploratory field and laboratory experiments, and also conducted seasonal field surveys.

METHODS

Objective 1 – Potential EPT species in the Glen Canyon Dam tailwaters and CRE

We collected EPT distribution information for species potentially occurring in the Glen Canyon Dam tailwaters and GCE (Fig. 1) from multiple sources, including the National Museum of Natural History (the Smithsonian Institution) in Washington, DC (NMNH); the Monte L. Bean Life Sciences Museum at Brigham Young University in Provo, Utah (BYU); the University of Arizona in Tucson (UA); and the Museum of Northern Arizona (MNA; Appendix 1A). Specimen data collected by museum data-mining included the following (where available): (1) locality, (2) collection date, (3) elevation, (4) number of individuals, (5) sex of individuals, and (6) flight dates. We also compiled information from the literature, including Woodbury et al. (1959), Knight and Gaufin (1966), Stewart et al. (1974), Flint and Hermann (1976), Baumann and Olson (1984), Hermann et al. (1986), Kondratieff et al. (1990), Moulton et al. (1994), Lugo-Ortiz and McCafferty (1995), Nelson and Baumann (2001), Kondratieff and Baumann (2002), Brammer and MacDonald (2003), Blinn and Ruiter (2006, 2009), Gunnison County (2011), McCafferty et al. (2012), Rasmussen and Morse (2016), and DeWalt et al. (2017). Tolerance values (TV) for each EPT genus were compiled from Appendix 1B in Barbour et al. (1999). Barbour et al. (1999) did not directly provide TVs for EPT in the Southwest. Therefore, for genera with multiple TV values across the USA (i.e., with different values in at least two of the five national regions), we calculated the median TV. Where species-specific information was provided (rather than genus-level information), the median species value was taken (across all species and all regions). In addition to TV, we also identified functional feeding groups (FFG) for each genus (Barbour et al. 1999, Merritt et al. 2008).

Biogeographic affinity and occurrence data also were compiled from state-based range data compiled from the above EPT literature sources. From reported state-based distribution data collected above, we evaluated the likelihood of each EPT species occurrence in three regions: (1) GCE, (2) Upper Colorado River Basin (UCRB), and (3) Lower Colorado River Basin (LCRB; Fig. 1). Species reported from one of these regions was considered detected. If a species was not detected in museum data-mining or the literature, but was reported from all Four Corners states, or in UT and CO, AZ and CO and/or WY, or NM and UT, its occurrence in the UCRB was considered likely. If it was reported in NM and AZ, or in NM and CA and/or NV, it was considered likely to exist in the LCRB.

Water quality data for EPT species have been erratically documented in the Southwest. Some general data exist in Barbour et al. (1999), in Merritt et al. (2008), and in Springs Online (the Springs Stewardship Institute's online database; springsdata.org), as well as from the U.S. Geological Survey's (USGS) National Water Information System, and the Environmental Protection Agency's (EPA) STORET website (EPA 2016, SSI 2016, USGS 2016). We focused on the following field water quality variables due to their importance to survivorship of BMI and the relatively high frequency of data reporting: pH, dissolved oxygen (DO) concentration, total alkalinity, water temperature, and specific conductance (SC). We also calculated ranges and medians for each of these water quality parameters from Lees Ferry for comparative purposes.

Objective 2 – Investigate BHA distribution and seasonal change in the Colorado River in the Glen Canyon Dam tailwaters

Study Area

The focal area for the field investigations of BHA included lower Lake Powell (the forebay of the dam) and the Glen Canyon Dam tailwaters from the base of the dam to approximately 2 kilometers below Lees Ferry (including the mouth of the Paria River; Fig. 2). Twelve transects were established for field measurements, including three sites in lower Lake Powell, six sites in the Glen Canyon reach from Glen Canyon Dam downstream to Lees Ferry, and three sites downstream from Lees Ferry to river kilometer 2 (Rkm 2).

The study area encompassed approximately 30 km from lower Lake Powell to just below Rkm 2. Riparian vegetation in this reach consisted primarily of saltcedar (*Tamarix* spp.), willow (*Salix gooddingii*), and cottonwood (*Populus fremontii*; Ralston 2005, Palmquist et al. 2017). Benthic aquatic vegetation consisted of dense stands of *Chara* nr. *vulgaris*, horned pondweed (*Zannichellia palustris*), and limited *Cladophora glomerata* cover. Benthic invertebrates of the reach are dominated by *Potamopyrgus antipodarum* New Zealand mud snails, *Dreissina buggensis* quagga mussels (in early stages of colonization), *Physa* snails, *Gammarus lacustris* scud, Turbellaria flatworms, Oligochaeta and lumbricid segmented worms, several

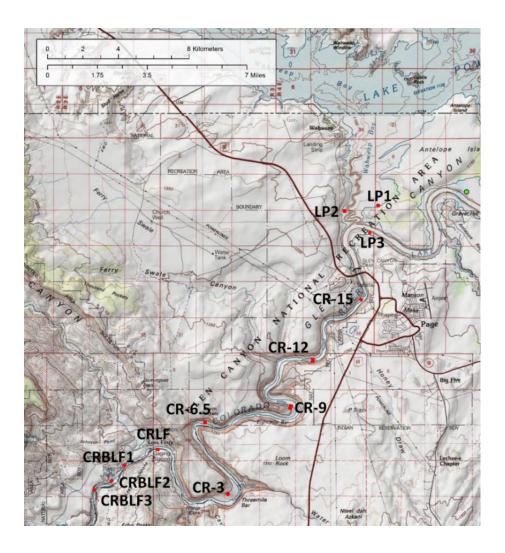


Fig. 2 Map of study area showing transect establishment sites, including Lake Powell sites (LP1, LP2, LP3), Colorado River sites between Glen Canyon Dam and Lees Ferry (CR-15, CR-12, CR-9, CR-6.5, CR-3), Lees Ferry (CRLF), and Colorado River sites below Lees Ferry (CRBLF1, CRBLF2, CRBLF3). All sites are located in the Glen Canyon National Recreation Area.

Chironomidae midge species, and *Simulium arcticum* blackflies (Stevens et al. 1997, 1998; Sublette et al. 1998, Kennedy et al. 2016).

Reservoir Stage and Dam Releases During the Study

Lake Powell stage changes and Glen Canyon Dam releases during the study period were seasonally normal in relation to reservoir inflows and operational requirements of the AMP 1996 and LTEMP 2017 Records of Decision (Fig. 3a; Appendix 2). As usual, Lake Powell reservoir pool stage peaked in June-July and gradually decreased throughout the subsequent summer, fall, and winter months, reaching nearly its lowest point during our March transect sampling period. Downstream, river flows varied during the low-release springtime and autumn months from 142 - 396 m³/s, with daily stage changes \leq 227 m³/s, and bimodal (summer and winter) higher flows, with a maximum of 510 m³/s (Fig. 3b). Daily flow fluctuations created

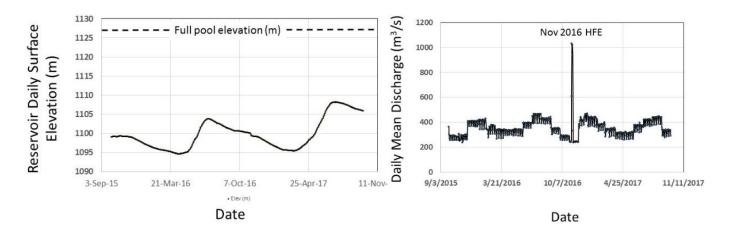


Fig. 3: a - left) Lake Powell Reservoir stage elevations, and b - right) Colorado River flow at Lees Ferry from 1 October 2015 through 30 September 2017. HFE – high flow experiment. Data from the U.S. Bureau of Reclamation (<u>https://www.usbr.gov/uc/water/crsp/cs/gcd.html</u>) and the Geological Survey (<u>https://waterdata.usgs.gov/nwis/inventory/</u>?site_no=09380000), respectively.

approximately 0.5 m of stage change at transects downstream from the dam, and the *in situ* fluctuating flow experiment.

Field Data Collection

Objective 2a – Transect Surveys: To determine and monitor the distribution and seasonal extent of BHA, we established 12 transects from lower Lake Powell through the Glen Canyon Dam tailwaters (Fig. 2). Three sites were established in lower Lake Powell, and nine sites were established within the Glen Canyon tailwaters (five upstream of Lees Ferry, one at Lees Ferry, and three below Lees Ferry). Each of the 12 transects was visited guarterly (summer, fall, winter, and spring from 2016 to 2017). A Sokkia C3₃₀ level, elevation rod, and a survey tape were used to measure elevation profiles (e.g., Fig. 4). Aspect was noted to ensure future repeatability and back sight elevation and distance were checked before and after each survey to ensure accuracy. Data were collected across the profile at abrupt changes in elevation or substrate. At each point, elevation and distance were measured and a pit was excavated to record substrate stratification and the presence and depth of BHA, as detected by conspicuous blackening of the sediment. Additional data recorded included vegetation type, water depth, and ecosystem characteristics (e.g., location within the riparian zone). Surveys were extended into the mainstream as far as wading safety would allow. Beyond the furthest survey point accessible on foot, a Petite Ponar dredge was deployed from the research boat to examine substrate conditions on the channel bed using a calibrated rope and rangefinder to determine depth and distance from the transect benchmark, respectively. Contents from the dredge were examined on site. Data were analyzed to determine whether BHA fluctuated seasonally or in response to daily and monthly stage fluctuations.



Fig. 4. Example of the BHA transect at CR-9. The pin flag indicates the upslope end of the transect, which continued into the channel. All measurements of BHA sediment were taken along this transect during each season, either on foot where permissible or using a Petite Ponar dredge in deeper sections.

Objective 2b – **Water and Sediment Geochemistry:** Field water chemistry data were measured at each of the 12 transects during each field trip using a YSI Professional Plus multiparameter instrument, calibrated daily for all parameters. We sampled water quality in the water column and in benthic sediments. BHA water samples were created using a 2:1 river water : BHA sediment slurry, which was settled for 10 min and then decanted. We collected the following water quality variables: temperature (°C), pH, dissolved oxygen (DO, mg/L), and oxidation-reduction potential (ORP, mV). We used t-tests (α = 0.05) to test for differences within parameters between the benthos and the water column.

In addition to field measurements, we collected water and benthic sediment samples for laboratory analysis at selected sites for the following components: Hg (water and sediments); nitrate (water and sediments) and nitrite (water); total P (water and sediments); sulfate (water); total U (water); total C and organic matter (sediments); and particle size (sediments). Laboratory water quality samples for the above variables except U were collected in summer and winter at CR-6.5R, CRLF, CRBLF3, and LP1 (Fig. 2), and during several points on the hydrograph of the November 2016 high flow experiment (HFE) at CRLF. Sampling dates were: 19 and 20 August 2016; 6-9, 11-12 November 2016 (HFE samples); and 7-8 March 2017. Water samples for U analysis were collected in BHA sediments in winter at CR-12, LP1, and CRBLF3. Water and sediment samples were kept on ice at ~4°C until analysis. Laboratory water quality analyses were conducted by Nortest Analytical Lab (Flagstaff, AZ) and by Pennsylvania State University Agricultural Analytical Services Lab (State College, PA).

Objective 2c – Macroinvertebrate Experiments: To test for effects of BHA on freshwater macroinvertebrates, we completed two exploratory and one detailed laboratory bioassay experiments using macroinvertebrate taxa from in and around the study area. The exploratory experimental designs used three concentrations of BHA water: 100% clean (tap) water, 1 clean: 1 BHA water, and 100% BHA water. The waters used in the experiment were decanted and acclimated for 48 hours prior to initiation of the bioassay. One hundred mL of water were added to plastic cups along with one macroinvertebrate per cup.

The initial exploratory bioassay was conducted for four days in July 2016 using damselflies (Odonata: Zygoptera - Coenagrionidae, 4/treatment), dragonflies (Odonata: Anisoptera-Libellulidae, 2/treatment), mayflies (Ephemeroptera-Baetidae, 2/treatment), and backswimmers (Hemiptera-Notonectidae, *Notonecta*, 3/treatment; Fig. 5) all collected from the Glen Canyon Reach. Percent mortality within each of the four taxa in each treatment was measured at the end of the experiment. Analysis of variance (ANOVA) was used to analyze differences in treatment (BHA concentration) effect using each species as an independent trial ($\alpha = 0.05$). The second exploratory bioassay focused on responses of two taxa in September 2016: coenagrionid damselflies (10/treatment) and *Gammarus lacustris* (7/treatment; Fig. 6). This experiment lasted 10 days. Percent mortality within each of the two groups in each treatment was measured at the end of the end of the experiment.

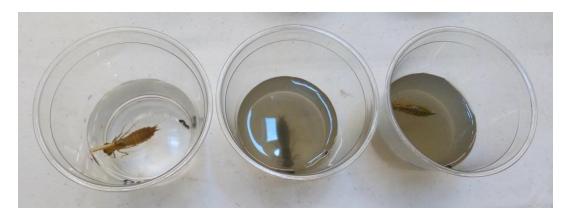


Fig. 5: The Anisoptera pilot macroinvertebrate bioassay showing the three treatments (left to right): clean tap water, 1 clean: 1 BHA water, and 100% BHA water. Only the dragonfly trials are illustrated.

Based on results from the first two bioassays, we conducted a third, more detailed bioassay by adding 50 mL of BHA sediment into 100 mL of Colorado River BHA water, compared against controls containing only Colorado River water (Fig. 7). *Heptagenia* sp. mayflies were collected from Oak Creek in Coconino County, AZ in March 2017, and one individual was added to each chamber (n = 30 of each treatment). Survivorship was measured after the first hour, and then each subsequent 12 hr for 8 days. Water quality (temperature, pH, conductivity, and DO concentration) were measured daily throughout the experiment. We performed a regression analysis on the survivorship data and analyzed the regression slopes in the experimental and control groups using a t-test (Zar 1984) to test for significant differences in

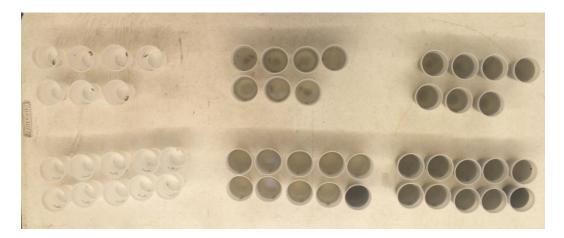


Fig. 6: The second laboratory macroinvertebrate bioassay used damselflies (top; 7/treatment) and amphipods (bottom; 10/treatment) among three water treatments (from left to right): clean distilled water, 1 distilled: 1 BHA water, and 100% BHA water.



Fig. 7: *Heptagenia* sp. larva, a common cool-water stream mayfly species throughout northern Arizona, but not occurring in the Colorado River mainstream. A laboratory bioassay experiment (experiment number 3) was conducted on this species to test the impacts of BHA sediment. A total of 50 mL of BHA sediment was added into 100 mL of Colorado River water, and mortality was compared against that in controls containing only Colorado River water over a 10 day period. mortality rate between groups. We also calculated the point at which 50% of the population perished (LD_{50})

Objective 2d – Determine the effects of simulated and high flow disturbance on BHA development: A field experiment was conducted to evaluate the impacts of mechanical disturbance on BHA development and BMI responses. We experimentally disturbed three 9 m² benthic plots at CR-6.5 (a low-velocity site) and at CR -9R (a high velocity cobble bar site) in August 2016 (Fig. 8). Both sites were strongly dominated by BHA, with the channel bed entirely black at the low velocity site, and the underside and deeper cobbles and gravels blackened at the high velocity site. During Sunday morning low-water releases, we used garden rakes to rigorously disturb benthic sediments 1 m downslope from the summertime minimum release stage. We monitored BHA discoloration on the treated plots for three months. At the lowvelocity site, we conducted a bioassay by anchoring experimental chambers (mesh bags, n = 3) on the floor of the channel. Each bag contained one *Gammarus*, one *Potamopyrgus*, and one larval baetid. Bags were monitored for mortality for 10 d.



Task 2e: Scouring Disturbance

Exptl. disturbance 3 reps each of 3 x 3 m patches of cobble versus sand: Effects lasted visually >60 days

No detectable benefits to BMI experimentally placed in treated vs. non-treated patches

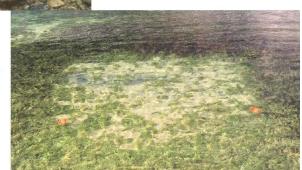


Fig. 8: Experimental mechanical disturbance of benthic, low velocity, fine sediment habitat at CR -6.5R (lower right), and coarse (cobble over gravel) high velocity habitat at CR -9R (upper left) in August 2016. Photographs were taken 12 hr after these sites were disturbed, and reveal rapid development of the buff-colored sand oxidized barrier layer in both settings.

A 96 hr-long, 1048 m³/s HFE was conducted through Glen Canyon Dam from 7-12 November 2016 (Fig. 3b). The HFE provided the opportunity to document changes in BHA distribution resulting from high flows. Prior to the HFE (November 1-6), transect surveys were completed at all 12 study sites. Water quality was measured at Lees Ferry during the HFE. In December 2016, following the HFE, transects were re-surveyed, water quality was remeasured, and pre- and post-HFE BHA distribution and depth were compared on each transect.

Objective 2e - Determine the conditions for, and developmental timing of BHA: We

investigated the time and factors influencing BHA development through field and laboratory experiments. In the field experiment, twelve 20 L plastic chambers were filled with clean (non-BHA) Colorado River sand and water at CR -6,5R. Six chambers were placed in a floating platform (Fig. 9) that maintained ambient river water temperature and were not overtopped by wave action or river flow fluctuations. Six other chambers were set into the shoreline at the middle stage elevation, with 8 cm of lip exposed, and were regularly inundated by waves and fluctuating flows. One L of living *Chara* algae was added to three chambers in each treatment. The stationary shoreline chambers containing *Chara* were covered with 1 mm mesh to prevent the algae from dispersing during inundation. The experiment began on 17 August 2016 and was completed on 23 September 2016. Water quality (temperature, DO, ORP, and pH) and the presence of BHA development (determined visually by blackening of the sediment) were measured initially and at the end of the experiment.

In order to determine how temperature, aeration, and presence of *Chara* interactively affect the development rate of BHA, we conducted a laboratory experiment using three 204-L tanks into which smaller chambers containing clean Lees Ferry sand were placed (Figs. 10, 11).

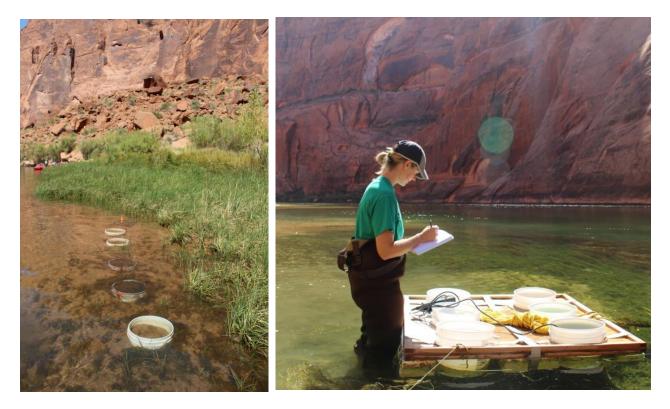


Fig. 9: BHA development field experiments. Floating chambers (right) were used to eliminate fluctuating flow impacts on BHA development, against controls in shoreline chambers (left) that were subject to daily flow fluctuations, and boat wake and wind wave impacts. Three of the six chambers in each treatment contained *Chara*, while the remaining three contained only non-BHA shoreline sand and mainstream water. Water quality measurements and presence of BHA were measured initially and at 38 days during the course of the experiment.



Fig. 10: Laboratory microcosm experiment: the three large chambers contain water baths maintained at each of (right to left) 5°C, 12°C, and 20°C. A total of 32 1-L microcosm chambers were placed together in a large containers, and each microcosm contained 1:1 volumes of clean Colorado River sand and water. Sixteen containers in each temperature treatment contained *Chara*, half (eight) of which were aerated using aquarium pumps and hosing.



Fig. 11: Laboratory microcosm experimental 1 L chambers at the conclusion of the 10-day experiment (Fig. 10). Depth of BHA development and sediment color was measured in each of the treatment combinations.

The three large 204-L tanks were each maintained at constant temperatures of 5°C, 12°C, and 20°C, spanning the range of water temperatures in the Glen Canyon reach. Each temperature bath contained thirty-two 1.0 L chambers (a total of 96 individual chambers), each of which were filled with 450 mL of dry buff (non-BHA) medium Colorado River sand and 450 mL of Colorado River water collected from Lees Ferry. Eight treatments in each thermal bath chambers contained *Chara*, *Chara* + aeration, aeration, or neither treatment. Aeration was provided using aquarium pumps and hoses (Fig. 10). Each temperature bath was placed under a 120-watt grow light simulating 8 hr of direct sunlight, and laboratory temperatures fluctuated little during the 10 d experiment.

Water quality parameters (pH, DO, ORP, and temperature) were measured initially (day 0) and on days three, six, and 10. On day 10, each 1 L chamber was removed from the temperature bath and examined for BHA development (Fig. 11). Depth of BHA development in the sediment was measured, and a Munsell Soil Color Chart was used to measure color differences among the treatments. We used a three-factor ANOVA followed by a Tukey posthoc test to determine differences within and among the three factors (temperature, *Chara* presence, and aeration), with $\alpha = 0.05$ for significance testing.

RESULTS AND DISCUSSION

OBJECTIVE 1 – EPT SPECIES OF THE COLORADO RIVER

The list of actual or potentially occurring EPT species in the Grand Canyon ecoregion, the drainage basin of Grand Canyon, includes at least 382 taxa, including 104 Ephemeroptera taxa among 41 genera in 10 families; 95 Plecoptera species among 44 genera in 8 families; and 185 Trichoptera species among 64 genera in 21 families (Appendix 1A). In Grand Canyon, we report a possible or detected total of 18 Ephemeroptera species among 15 genera in 6 families; 6 Plecoptera species among 5 genera in 3 families; and 45 Trichoptera species among 25 genera in 15 families. Thus, with the exception of Plecoptera, the existing EPT fauna in the GCE and Grand Canyon is relatively robust. While the diversity of EPT in tributary streams is relatively high, it remains depauperate in the Colorado River mainstream, with *Rhyacophila coloradensis* and one or more hydroptilids the only caddisfly species regularly encountered in the CRE. Those species may be useful as indicator species; however, the EPT fauna in the Colorado River mainstream remains remarkably depauperate, as noted by many previous researchers.

Analyses of water quality requirements, habitat, and feeding strategies (where known) indicate that most of the EPT species reported or potentially occurring in the GCE and GC appear to be capable of existing in some portion of the CRE mainstream under conditions of low flow variability and high water transparency (Appendix 1B). However, all Plecoptera except 3 species of Perlidae (*Hesperoperla* and *Isoperla*) occur at elevations far above that of the mainstream, and at least 101 GCE Trichoptera species (55%) occur at elevations above 1200 m and therefore at higher elevations than the mainstream. Although water temperatures are generally cooler at higher elevations, other water quality variables, such as conductance, often differ substantially from those in the CRE (e.g., Ledbetter et al. 2016). In addition, CRE suspended sediment loads, thermal constancy, benthic habitat structure, and daily flow fluctuations also may limit EPT colonization (Cross et al. 2013, Kennedy et al. 2016). Understanding the influences of those and other factors on individual EPT species life histories

and potential for colonization success in the Glen Canyon tailwaters reach will require detailed investigations. Our data indicate that factors other than water quality limit EPT colonization and survival in the mainstream Colorado River downstream from Glen Canyon Dam.

OBJECTIVE 2 – BHA DISTRIBUTION AND DEVELOPMENT (APPENDIX 2)

Objective 2a – Transect Surveys. Quarterly surveys along each transect revealed that BHA was commonplace throughout the Glen Canyon Dam forebay and in the tailwaters between the dam and Lees Ferry (Table 1, Fig. 3). BHA was apparently ubiquitous in sand and silt sediment deposits below the annual low water stage of Lake Powell reservoir at sites dominated by aquatic macrophytic vegetation and with extensive buried shoreline organic matter. Mainstream tailwaters flows varied daily and monthly during the study duration, varying from low of near 142 m³/s to nearly 510 m³/s during July and August of both 2016 and 2017 (Fig. 3b). BHA dominated all Glen Canyon reach transects from Glen Canyon Dam downstream to Lees Ferry, but it occurred rarely and episodically at the three sites located downstream from Lees Ferry to the Paria Beach (downstream from the Paria River confluence). BHA development generally advanced shoreward during the summer high flow period, and retracted to the lower low water stage in winter.

Transect	Summer: 14- 15 Aug 2016	Fall: 14-15 Dec 2016	Winter: 7-9 Mar 2017	Spring: 4-5 May 2017	
LP-1 Elev of BH (m)	1101.75*	1098.42	1095.11	none	
LP-2 Elev of BH (m)	1101.75	1100.81	1095.27	none	
LP-3 Elev of BH (m)	none	none	none	none	
CR -15R Distance from BM (m)	7.62	6.37	8.69	9.36	
CR -12R Distance from BM (m)	14.63	11.52	14.81	14.02	
CR -9R Distance from BM (m)	24.08		29.69	29.14	
CR -6.5R Distance from BM (m)	17.37	14.63	17.18	17.37	
CR -3R Distance from BM (m)	3.05	9.45	6.10	8.02	
LF R Distance from BM (m)	4.88	4.43	3.11	4.57	
BLF1 R Distance from BM (m)	8.23	12.19	none	none	
BLF2 R Distance from BM (m)	none	15.24	17.77	11.34	
BLF3 R Distance from BM (m)	none	7.01	6.37	5.23	

Table 1: Elevation of BHA (m) in Lake Powell (LP) or distance (m) from the benchmark to the nearest expression of BHA in the tailwaters, 2016-2017. * Estimated value.

BHA developed strongly in lentic settings floored with fine sediments below the lowest seasonal low water stage in the reservoir and in the river upstream from the Paria River confluence, particularly in settings dominated by the presence of dense benthic macrophyte cover (*Chara* sp., *Zanichellia* sp., other macrophytes). BHA extended to >30 cm depth in lentic settings, and >15 cm in cobble bar settings. Less BHA was detected in the mainstream thalweg, suggesting that PAR limitation with higher velocities and reduced macrophyte cover limited

BHA development. Limited but detectable BHA development occurred beneath gravels and cobbles in cobble bar habitats (e.g., CR -9R) upstream from the Paria River confluence, but not downstream in Grand Canyon.

BHA distribution varied seasonally in the reservoir and to a lesser extent in the tailwaters, developing relatively gradually following prolonged summertime inundation, and occurring at inundated higher stage elevations during the late summer and autumn months. BHA dissipated when reservoir and downstream river shoreline stages decreased in elevation for more than 12 hr during seasonal dewatering (e.g., late autumn; Table 1; Fig. 3). Although seasonal shoreline shifts in BHA were erratic, perhaps due to HFE impacts, BHA advanced shoreward at Lees Ferry by 1.49 m from summer to fall, 2016, and then retreated with low late winter and springtime flows. The development of BHA at Site CR-3 likely was due to the dense wetland vegetation and nearly constant inundation in the shallow return current channel depression there. Overall, the extent of BHA development along shorelines was greatest during early autumn, occurring further upslope during fall, and dissipating during winter and springtime.

BHA was not regularly detected at any of the three sites below Lees Ferry during the summer sampling period. However, traces of BHA were found among the cobbles at CRBLF1 and in shoreline sand deposits at CRBLF2. The furthest downstream site, CRBLF3, developed considerable amounts of BHA to >30 cm depth following Paria River flooding and deposition of extensive quantities of organic matter. However, those sediments and organic deposits were flushed from that eddy during the 2016 HFE, and BHA did not re-develop during the 2017 winter and springtime periods when benthic macrophyte production was low. Farther downstream, BHA was rarely detected in a few low-velocity tributary mud deposits during several river trips in 2016 and 2017. BHA development in the CRE downstream in Grand Canyon and its perennial tributaries is extremely limited, likely due to the rarity of aquatic macrophyte cover, high dissolved oxygen concentrations, and more dynamic sediment transport and flow regimes.

Objective 2b – **Water Column and BHA Sediment Geochemistry.** Field water chemistry measurements revealed no strongly significant differences (p > 0.05) between any of the parameters on the benthic surface versus the water column (Table 2). However, several variables of interest are discussed below.

Heavy Metals: Laboratory water quality analyses indicated little detectable mercury (Hg) or uranium (U) mobilization in BHA-dominated sediments, with nearly all samples below minimum detectable levels, except for sediments from Lees Ferry and CR-12. At Lees Ferry, contrary to our expectations, the Hg concentration was 5 μ g/L in BHA sediments, but was higher (7 μ g/L) in non-BHA sediments. The EPA has established a maximum contamination limit (MCL) for Hg in drinking water of 2 μ g/L (https://www.epa.gov/wqc), while the World Health Organization limit is 6 μ g/L. For acute (maximum) and chronic (continuous) freshwater aquatic systems, the EPA has established a limit of 1.4 and 0.77 μ g Hg/L, respectively. Our field sediment values at Lees Ferry were 2.5- to 3.5-fold higher than EPA MCL for drinking water, and were slightly higher than the World Health Organization's drinking water MCL. Those sediments were 3.6- to 5-fold higher than acute levels, and 6.5- to 9.1-fold higher than chronic exposure in fresh waters.

		Temp (°C)	DO (mg/L)		ORP (mV)	Hg (mg/L)	Nitrate (mg/L)	Nitrite (mg/L)	U (mg/L)	Mn (mg/L)	U (mg/L)2	Total C (%)	Mn (mg/L)2	Alkalinity (mg/L)	Total P (mg/L)	Sulfate (mg/L)	Sand	Silt	Clay	Organic Matter (%)
Site	Sample Type	Ter	DO	Нq	ORP	Hg (Nitr	Nitr	n (n	ЧИ	u) U	Tota	μ	Alka	Tota	Sulf	% Si	% Si	% CI	Org
BLF1	Water column	14.4	10.4	8.27	225															
BLF2	Water column	15.1	9.6	8.31	219															
BLF3	Water BHA									0.24			0.24	169		215.5				
BLF3	Water column	13.5	8.8	8.16	188		0.32							143	0.84	200.8				
BLF3	Water-upper well																			
BLF3	Sed. Non-BHA						8.15					0.37			61.44		97.9	0.6	1.5	0.00
BLF3	Sed. BHA						3.07					0.88			73.55	189.0	88.7	8.4	2.9	0.04
CR -12R	Water BHA	12.8	7.2	7.95	238				0.0188		0.02									
CR -12R	Water column	12.6	7.1	7.95	238															
CR -12R	Water-lower well	15.3	2.9	7.43	248															
CR -12R	Water-upper well																			
CR -15R	Water BHA	13.9	8.4	8.10	76															
CR -15R	Water column	13.9	8.0	8.05	74															
CR -15R	Water-lower well	16.8	2.2	7.28	227															
CR -15R	Water-upper well	20.2	1.2	7.61	222															
CR -3R	Water BHA	14.7	9.0	8.25	37															
CR -3R	Water column	13.9	9.9	8.21	99															
CR -3R	Water-upper well																			
CR -6.5R	Water BHA	12.7	7.3	7.84	157		0.27							166	3.14	206.3				
CR -6.5R	Water column	12.3	7.1	7.77	156		0.39			0.01			0.01	145		203.4				
CR -6.5R	Water-lower well	15.1	2.9	7.36	310															
CR -6.5R	Water-upper well																			
CR -6.5R	Sed. Non-BHA						7.54					0.26			105.46		95.3	2.2	2.5	0.01
CR -6.5R	Sed. BHA						31.50					0.44			91.15		93.1	3.3	3.6	0.08
CR -9R	Water BHA	16.6	6.9	7.79	274															
CR -9R	Water column	13.1	8.6	7.98	281															

Table 2: Average field and laboratory sediment (Sed.) and water geochemistry in paired wells (flowing transects only), the shallow hyporheic zone, and the water column at each of the study sites, based on seasonal sampling in 2016-2017.

Site	Sample Type	Temp (°C)	DO (mg/L)	Hd	ORP (mV)	Hg (mg/L)	Nitrate (mg/L)	Nitrite (mg/L)	U (mg/L)	Mn (mg/L)	U (mg/L)2	Total C (%)	Mn (mg/L)2	Alkalinity (mg/L)	Total P (mg/L)	Sulfate (mg/L)	% Sand	% Silt	% Clay	Organic Matter (%)
LF	Water BHA	12.7	7.6	7.90	114		0.31			0.02			0.02	354	1.97	192.6				
LF	Water column	12.5	7.7	7.92	111		0.31							144	0.06	198.7				
LF	Sed. BHA					0.005	5.30					1.08			212.47		77.2	16.5	6.3	0.71
LF	Sed. Non-BHA					0.007	12.10					3.88			297.91		74.2	17.7	8.1	4.99
LP1	Water BHA	22.3	3.6	8.23	136										2.24	168.0				
LP1	Water column	16.5	8.7	8.50	87											192.0				
LP1	Water BHA	26.5	3.5	8.02	188		4.13					0.03					98.0	0.6	1.4	0.00
LP1	Water column	28.1	6.8	8.26	183		5.55					0.30			30.50		93.4	2.9	3.7	0.04
LP2	Water BHA									0.05			0.05	205		186.6				
LP2	Water column						0.20							125		172.3				
LP2	Water BHA	23.3	5.5	8.43	145															
LP2	Water column	20.8	6.9	8.38	133															
LP3	Water BHA	22.9	5.9	8.30	160															
LP3	Water column	20.7	7.3	8.37	140															

Two of the three substrate samples (LP1 and CRBLF3) analyzed for uranium revealed U concentrations at or below reported detection limit (0.01 mg/L). However, the CR-12 sample contained 0.019 mg/L U, slightly above the reported detection limit but below the EPA's maximum contamination limit (MCL) for drinking water (MCL = 0.03 mg/L).

Nutrients: Our water quality sampling revealed no detectable release of nitrate, nitrite, or sulfate from BHA sediments. However, during the summer 2016 sampling period, we detected increased total P concentration from water extracted from BHA sediments. BHA sediment samples revealed slightly elevated total P, nitrate, total C, and organic matter compared to non-BHA sediments at sites CR-6.5 and LP1. The opposite pattern was documented at CRLF, where BHA sediments contained lower concentrations of total P, nitrate, total C, and organic matter compared to non-BHA sediments at sites compared to non-BHA sediment samples.

Conclusions: BHA impacts on Colorado River water quality appear to be minor at the scale of individual sampling points, based on laboratory analyses of field water column and BHA sediments. Our results appear to be consonant with the findings of Wildman et al. (2010) in Lake Powell Reservoir, who reported minor seasonal releases of U from benthic sediments. Low levels of geochemical activity in BHA sediments and at the sediment-water interface may nonetheless result in cumulative, long-term, element-specific release or sequestration downstream. In addition, potential releases of P and N from BHA sediments during HFEs may increase tailwater productivity. The release of toxic compounds from benthic sediments under normal and HFE flows may warrant further study.

Objective 2c – Macroinvertebrate Experiments. Between two separate macroinvertebrate experiments, we documented a small degree of negative effect of BHA water on various freshwater species. Our initial experiment using damselflies, dragonflies, and mayflies collected in off-channel pools in the Glen Canyon Reach, resulted in a significant treatment effect when all species were combined into one analysis, with each species representing an independent trial ($F_{2,12} = 5.39$, p = 0.021). We documented greater percent mortality in 100% BHA water compared to the 50:50 dilution treatment (p = 0.023), and marginal differences between the 100% BHA and clean tap water treatments (p = 0.072). These results were interesting, but the lack of resolution was due to the use of different taxa in the comparison, as well as the relatively low statistical power of the experiment.

The second bioassay experiment tested BHA water on damselflies and amphipod mortality. We found 0% mortality in both BHA and 50:50 BHA waters, but found greatest mortality among both taxa (10% in damselflies and 60% in amphipods) in distilled water. These results likely were due to the highly tolerant status of the two aquatic macroinvertebrate taxa, but their intolerance of low solute concentrations in the distilled water treatment.

The third, more tightly focused bioassay experiment involved subjecting *Heptagenia* mayfly larvae to 10-day treatments of BHA sediment with Colorado River water against controls without BHA sediments. This experiment generated significant negative effects on *Heptagenia* survival in the BHA treatment ($t_{0.05, 19} = 2.213$, p = 0.039; Fig. 12). By the fifth day, all mayflies in the BHA treatment had died, whereas only 40% of *Heptagenia* mayflies in the control treatment had perished. Water quality differed as well, with electrical conductivity and DO

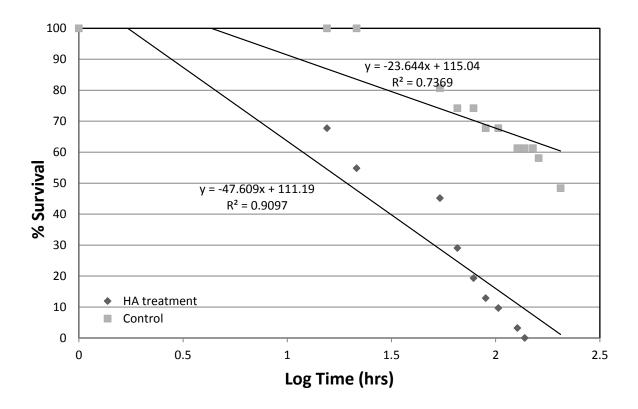
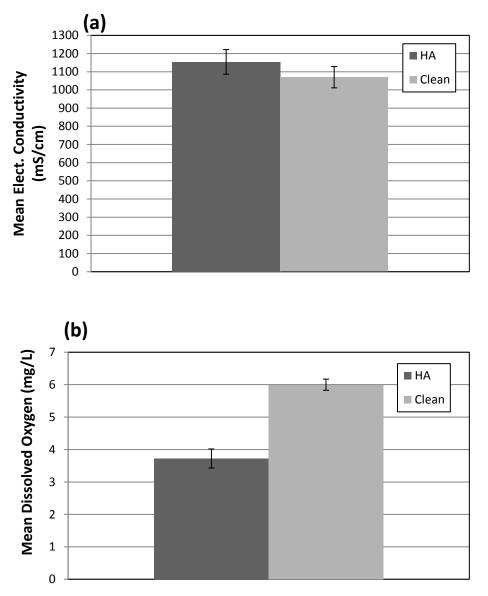
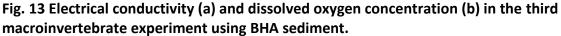


Fig. 12 Percent survival of *Heptagenia* sp. mayflies in the presence of BHA sediments ("BHA treatment") or controls. The x-axis is log-scale of the time over which the experiment took place.

differing between the two treatments (Fig. 12). The 50 percent mortality rate (LD_{50}) in the BHA treatment occurred at 18.3 hrs after the start of the experiment, and LD_{50} in the control group was expected to occur at 562 hrs after the start of the experiment (the experiment was discontinued after 10 days). Heptagenia mayfly larvae are lotic species, and are more sensitive to poor water quality than are damselflies or amphipods (i.e., the second macroinvertebrate experiment described above). No mayfly species are presently found in the mainstream of the Colorado River in Glen or Grand Canyons, although mayflies are abundant in springs and tributaries throughout the CRE. The high degree of mortality caused by BHA fine organic sediments may be one factor contributing to this absence, particularly in lentic settings where the extremely fine particle size of BHA sediments may physically obstruct mayfly respiration. Thus, this physical impact of macrophytic plant decomposition may more strongly limit mayfly presence than does the geochemistry of the BHA sediments. The first two experiments using only BHA water did not generate distinctive impacts on test organisms. The existing benthic fauna in the tailwaters reach are largely non-native (i.e., New Zealand mudsnail, quagga mussel, Gammarus scud, lumbriculid worms, and possibly Physa snails) and all appear to be highly adapted to survival in BHA-dominated sediments and waters.





Objective 2d – Artificial Flood Disturbance: Bed disturbance was experimentally simulated by raking three 9 m² patches in lentic and cobble bar habitats in August 2016. The patches were initially black in color, but within 12 hr had undergone oxic transition. The lentic (sand-floored) channel setting at -6.5R developed a 1 cm buff sand surface layer that persisted until the onset of the November 2016 HFE (below). The -9R cobble floored channel similarly lost its black anoxic appearance, and remained visible for more than a month. Experimental caging of benthic macroinvertebrates onto treated and untreated benthic surfaces did not reveal clear differences in mortality, likely because the organisms used (*Gammarus, Physa* and New Zealand mud snails) are highly tolerant of BHA conditions.

The post-HFE transect surveys at CR-6.5 and CRLF indicated a minor recession of BHA distribution in the near-shore habitat as a result of artificial flooding (Table 2). Prior to the HFE, BHA was found 0.18 m (at LF) and 0.61 m (at CR-6.5) closer to shore than during the post-HFE period, indicating a small amount of scour and aeration of the sediment resulted from the HFE. This minor recession suggests that HFEs can have some effect on reducing the amount of HA. In addition, the HFE scoured some *Chara* cover and some of the decomposing macrophytic organic matter from the bed, resulting in development of a 1 cm-thick buffer layer of oxic sand throughout the tailwaters. This buffer layer persisted into the May transect survey in 2017.

Collectively, these results indicate that fine-scale or minor system-wide artificial disturbances can regenerate surficial oxic conditions on the bed that may last for biologically meaningful time periods. However, subsurface sediments beneath the buffer layer retained their blackened BHA character, and without further disturbance or implementation of sediment-disturbing actions, BHA gradually re-develops. Increased frequency, duration and magnitude of releases may exert longer-lasting impacts on BHA below Glen Canyon Dam, but further research is warranted into the specifics of HFEs and other flow management actions needed to mitigate or reverse BHA development.

Objective 2e – BHA Development Experiments. The field mesocosm experiment revealed significant development of BHA within the 38-day experimental period (Figs. 14, 15). BHA developed only in chambers into which *Chara* had been added. BHA development in that experiment occurred to equal degrees in both the floating and submerged treatments. Water quality was generally similar between the submerged and floating treatments over 38 days (Figs. 16, 17). The presence of *Chara* led to decreased dissolved oxygen concentration, pH, and ORP in both treatments, indicating that the presence of *Chara* is a strong driver of BHA.



Left Fig. 14, Right; Fig. 15, Left: Photographs of BHA sediment that developed *in situ* in stationary (left; stage-varying) and floating (right; stage-constant) treatments with *Chara* at the end of the 38-day field experiment. BHA developed equally in both treatments from the surface downwards, leaving oxic sediment (buff colored sand to the upper right) below it. Strands of *Chara* also can be observed in this photograph.

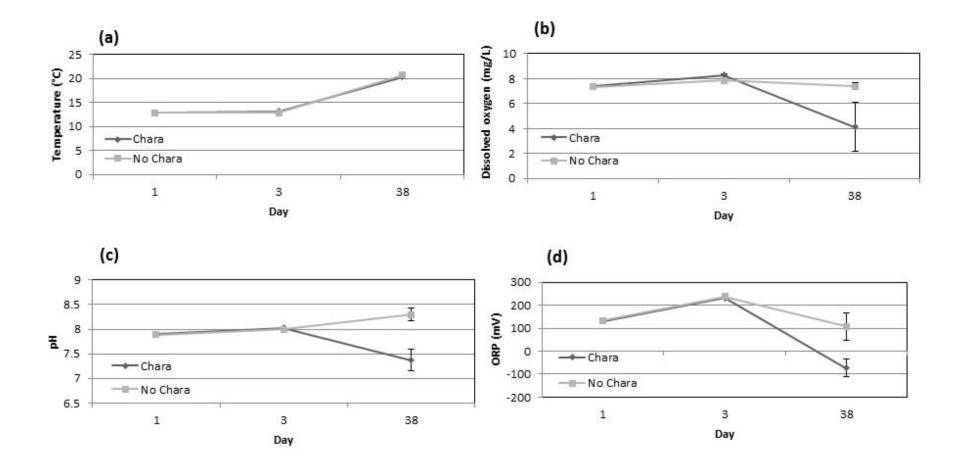


Fig. 16: Water quality variables in the stationary shoreline chambers during the 38-day field BHA development experiment: a) temperature, b) dissolved oxygen, c) pH, and d) oxidation-reduction potential (ORP). Error bars are standard error. N = 6 chambers, with 3 chambers containing *Chara*.

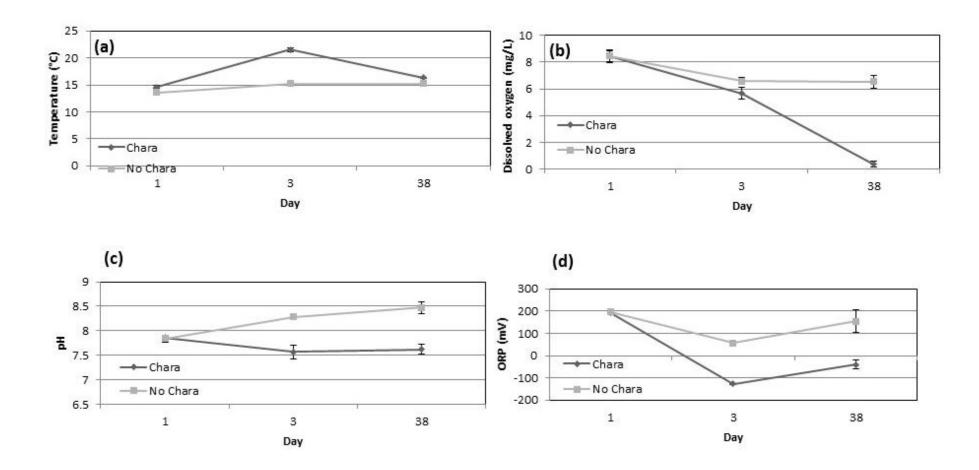


Fig. 17: Water quality variables from the floating chamber treatment in the 38-day field BHA development experiment: a) temperature, b) dissolved oxygen, c) pH, and d) oxidation-reduction potential (ORP). Error bars are standard error. N = 6 chambers, with 3 chambers containing *Chara*.

This initial field experiment emphasized the importance of macrophytic vegetation on dissolved oxygen concentration and BHA development, and suggested that the minor flow fluctuations (0.5 m/day) and boat wake wave actions do not greatly influence the BHA developmental process. More research into the effects of different algal taxa (e.g., *Chara* vs. *Cladophora;* Crayton and Sommerfield 1979, Ariosa et al. 2004) and vascular plants (e.g., *Zanichellia*) on BHA development is needed to determine the extent to which different plant taxa influence BHA development and the ecological integrity of the tailwaters.

The laboratory microcosm BHA development experiment provided significant insight into the effects of water temperature, dissolved oxygen concentration, and the presence of *Chara* on rates of BHA development on water quality. Water quality (pH and oxidation-reduction potential [ORP]) varied significantly over the course of that 10-day experiment (Figs. 18, 19). Presence of *Chara* led to decreased DO, pH, and ORP, particularly at higher temperatures and in the absence of aeration.

All three treatment variables in the laboratory microcosm experiment (presence of *Chara*, temperature, and aeration) significantly affected the development of BHA (measured as depth of darkened sediment at the end of the experiment): *Chara*: $F_{1,84} = 1244.75$, p < 0.001; temperature: $F_{2,84} = 12.77$, p < 0.001; aeration: $F_{1,84} = 21.96$, p < 0.001. No BHA development occurred in the absence of *Chara*, and sediment in all chambers with *Chara* present developed conspicuous BHA. This indicates *Chara* (and possibly other types of aquatic vegetation) is necessary for anoxia to develop in benthic sediments, supporting the hypothesis that this phenomenon is attributable to large quantities of decaying vegetation. In addition to presence of *Chara*, higher temperatures ($T_{20} > T_5$, p < 0.001; $T_{20} > T_{12}$, p = 0.037; $T_{12} > T_5$, p = 0.0341) and lack of aeration (non-aerated > aerated, p < 0.005) led to increased BHA development rates (Fig. 18).

In addition to effects of each variable independently, significant interactions occurred among all variables with respect to BHA development. Aeration in the presence of *Chara* significantly reduced BHA depth when compared to non-aerated treatments with *Chara* (p < 0.001). Thus increased aeration in the Glen Canyon Dam tailwaters may limit BHA development. However, BHA development still occurred even in the presence of aeration, albeit to a lesser extent. A three-way comparison showed that aeration at elevated temperatures can mitigate temperature effects, resulting in decreased BHA development (i.e., no significant differences were detected between aerated treatments with *Chara* present at 12°C or 20°C versus nonaerated treatments with *Chara* present at 5°C). This provides further evidence that aeration can significantly reduce BHA development. However, the difference between the low (5°C) and the middle (12°C) temperature treatment in the absence of aeration did not change BHA development rate. At higher temperatures (20°C), BHA rate of development significantly increased in the absence of aeration. The highest water temperature in our experiment generated the most rapid development of BHA.

In summary, the combination of warm water temperature, the presence of *Chara*, and the lack of aeration promoted the most rapid development of BHA in benthic sediments. Altering any of these three factors (i.e., decreasing water temperature, removing *Chara* biomass, or increasing aeration) or tandem combinations thereof, will likely retard BHA development.

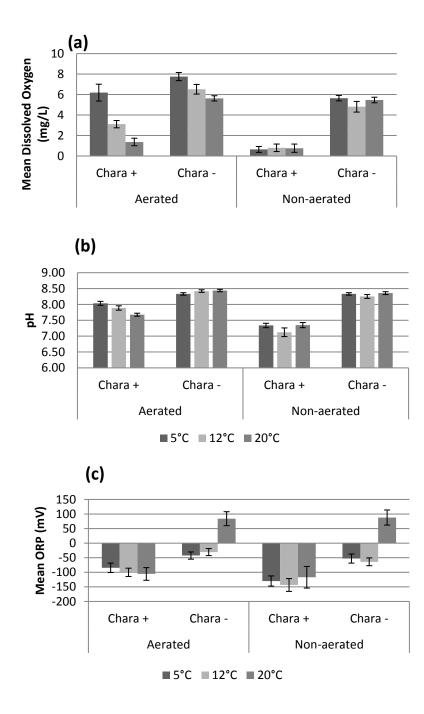


Fig. 18: Water quality parameters for each of the experimental treatments, measured three times over the 10-day experiment: (a) mean dissolved oxygen (mg/L), (b) mean pH, and (c) mean oxidation-reduction potential (mV). Values represent the mean measurement for each parameter over the course of the experiment. Error bars are standard error. Measurements were taken using a daily-calibrated YSI ProPlus multiparameter meter.

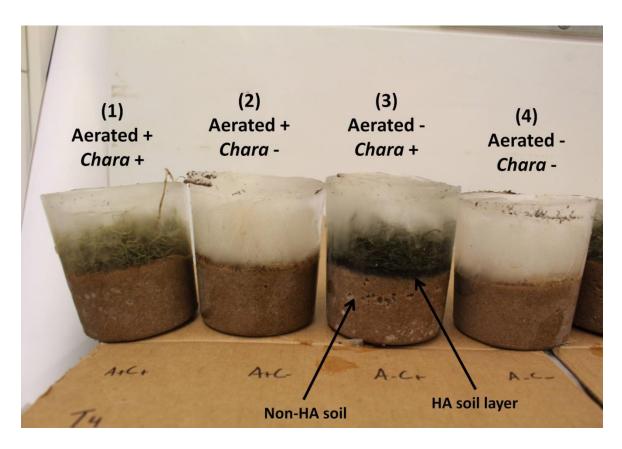


Fig. 19: Laboratory BHA development experiment after 10 days of treatments. Treatments (from left to right) are: 1) aerated, with *Chara*, 2) aerated, without *Chara*, 3) non-aerated, with *Chara*, and 4) non-aerated, without *Chara*. BHA is evident by the black color of surficial strata on top of buff-colored sand. Depth and color of the BHA and non-BHA sediments were measured at the conclusion of the experiment.

MANAGEMENT IMPLICATIONS AND RECOMMENDATIONS

Objective 1 – CRE EPT

We report that at least 382 EPT taxa have been documented or potentially could occur in the Grand Canyon ecoregion, which is the drainage basin of Grand Canyon. These EPT include 104 Ephemeroptera taxa among 41 genera in 10 families, 95 Plecoptera species among 44 genera in 8 families, and 185 Trichoptera species among 64 genera in 21 families (Appendix 1A). In Grand Canyon, we report a possible or detected total of 18 Ephemeroptera species among 15 genera in 6 families, 6 Plecoptera species among 5 genera in 3 families, and 45 Trichoptera species among 25 genera in 15 families. Thus, with the exception of Plecoptera, the existing EPT fauna in the GCE and Grand Canyon is relatively robust. However, as much previous research has demonstrated, the EPT fauna in the Colorado River mainstream remains remarkably depauperate.

This study provides a more complete understanding of the total diversity of EPT taxa that do, or potentially could be, expected to occur in the Colorado River downstream from Glen Canyon Dam. We did not find strong evidence to suggest the taxa potentially occurring in the

Glen and Grand Canyon reaches are or would be excluded by inhospitable water quality conditions in the dam-regulated tailwaters or downstream reaches: temperature, pH, SC, and DO concentration ranges appeared to be largely compatible within reported ranges for those taxa. However, except for Trichoptera, few data are available regarding species-specific impacts of suspended loads, seasonal temperature variation, or benthic habitat requirements (e.g., embeddedness, presence of BHA) for EPT taxa, all of which and life history constraints may strongly influence potential colonization success in the CRE.

Blinn and Ruiter (2009a) reported low Trichoptera species richness at elevations below 1000 m in Arizona, with dominance there by *Hydroptila arctia, H. icona, Octrotrichia inconspicua, O. logana, Oxythira arizona, Neotrichia olorina,* and *Smicridea fasciatella;* however, they commented that no Trichoptera species were useful as indicators of river ecosystem health below 1000 m elevation in Arizona due to excessive anthropogenic impacts on rivers and streams. In addition to the species they report, we found that *Neotrichia* spp., *Hydropsyche oslari,* and *Rhyacophila coloradensis* were regularly encountered in lower CRE reaches, as well as in perennial tributaries, and other species occur in desert springs in the region (a habitat not studied by Blinn and Ruiter 2009a,b; Appendix A). Furthermore, two caddisfly species cause nuisance outbreaks in the Parker and Davis reaches downstream from Hoover Dam: *Smidicrea fasciatella* and *Nectopsyche spilosa,* identified by Dr. Oliver S. Flint, Jr (NMNH). Our study integrates the distribution of those and other EPT taxa into the context of the regional fauna, and indicates that several species may be of interest in CRE foodbase management.

Modification of the above variables to enhance EPT presence in the mainstream all have been the subject of recent inquiry by river managers, but none of the possible measures have yet been implemented. Biotic and habitat relationships among EPT are complex (Haden et al. 1999, Griffith et al. 2001), and species-specific tests of the individual and interactive impacts of those variables would require detailed experimental investigation, as well as field tests. While notable investigations are being conducted on selected life history stages of several EPT taxa in the upper Colorado River basin (Miller, oral communication), application of those findings to management actions at Glen Canyon Dam or elsewhere in the lower Colorado River basin remain outstanding.

We conclude that while several too many EPT taxa could potentially exist in the Glen Canyon reach or in the downstream CRE, enhancing EPT diversity in those reaches is likely to require substantial changes in Glen Canyon Dam operations and perhaps in the diversity of fluvial habitats available. While, EPT diversity in the CRE could be greater than the present low levels, trade-off assessment among management options and the values and uncertainties of biological responses will require additional research. Water quality, with perhaps the exception of seasonal variation, does not appear to be a limiting factor in EPT distribution in the Colorado River below Glen Canyon Dam. The limiting mechanisms remain unclear, but fluctuating flows have been implicated as a mechanism that could reduce egg hatching success (Kennedy et al. 2016). Understanding river habitat requirements and dam operations impacts on individual taxon life histories and habitat requirements are presently poorly known for most western EPT taxa, and additional field research at tributaries, such as Tapeats Creek in Grand Canyon, as well as more detailed laboratory studies of physiology and life histories are needed to distinguish dam operation (flow fluctuation) impacts from other factors limiting EPT diversity in the highly regulated Colorado River in Glen and Grand Canyons.

Objective 2 – BHA

Investigation into the details of BHA distribution and development in Glen Canyon and Lake Powell revealed previously unrecognized patterns of ecosystem development that may be of interest in ecosystem management (Appendix 2). The vast majority of BHA-affected sediment was located in the lower stage elevations of Lake Powell and the Colorado River downstream to Lees Ferry. BHA was ubiquitous in those reaches, and was particularly prominent in lentic, sand-dominated sections of the channel where aquatic macrophytic vegetation was most dense, in accord with the findings of Banks et al. (2012). Cobble bars and benthic sediments in higher velocity sections of the channel contained somewhat less evidence of BHA presence. Depth of BHA ranged from one to >30 cm (the extent of submerged sediment we were able to excavate). The HFE in November 2016 caused BHA to recede slightly from the shoreline, but this response was minimal and relatively short-lived. The extent of BHA development documented in the Glen Canyon Dam tailwaters is considerable, with virtually all of the river bed downslope of the 200 m³/s stage (approximately 2 km² of the channel) affected by BHA. Due to difficulty in assessing the depth of BHA sediment at each transect site, the volume of BHA sediment in the Glen Canyon Dam tailwaters remains unknown. BHA was detected in the CRE downstream from the Paria River confluence ephemerally in fine (non-*Chara*) sediment deposits containing high concentrations of organic matter. However, such occurrences of BHA are erratic and short-lived in Grand Canyon.

The guality of water extracted from BHA and non-BHA sediments differed. In general, BHA sediments contained elevated total P, total C, and organic matter as compared to non-BHA sediments and the river water column. The concentrations of total P was slightly elevated during summer in BHA sediments as compared to non-BHA sediments, particularly downstream from the Paria River and perhaps in relation to differing concentrations of autochthonous vs. allochthonous organic production. Hg was detected in sandy sediments at Lees Ferry; however, contrary to expectations, slightly greater concentrations of Hg were found in non-BHA sediments there. The elevated Hg concentrations found in non-BHA sediments at Lees Ferry exceed the EPA's maximum contaminant level (MCL) for drinking water (MCL = 0.002 mg/L), and non-BHA sediments contained greater concentrations than the World Health Organization's drinking water guidelines (WHO guideline = 0.006 mg/L). Higher levels of Hg in CRLF sediments may reflect long-term human development and the historic use of the area immediately upstream from the Lees Ferry launch ramp (i.e., the Spencer mining operation, the historic trading post and settlement, and the former marina there). Overall, these data indicate that benthic anoxia is not mobilizing Hg at readily detectable levels at the micro-site scale; however, minor releases of Hg and U across the entire channel bed may be occurring and warrant continued monitoring.

We conceptually modeled this process to visually explain the causes of BHA development in the Glen Canyon Dam tailwaters (Fig. 20). BHA development in Lake Powell reservoir and in the Glen Canyon Dam tailwaters is strongly related to the presence of *Chara* nr. *vulgaris* and other benthic macrophyte taxa. *Chara* is known to influence aquatic ecological processes in lentic habitats in complex ways (Kufel and Kufel 2002). The dominance and role of *Chara* in

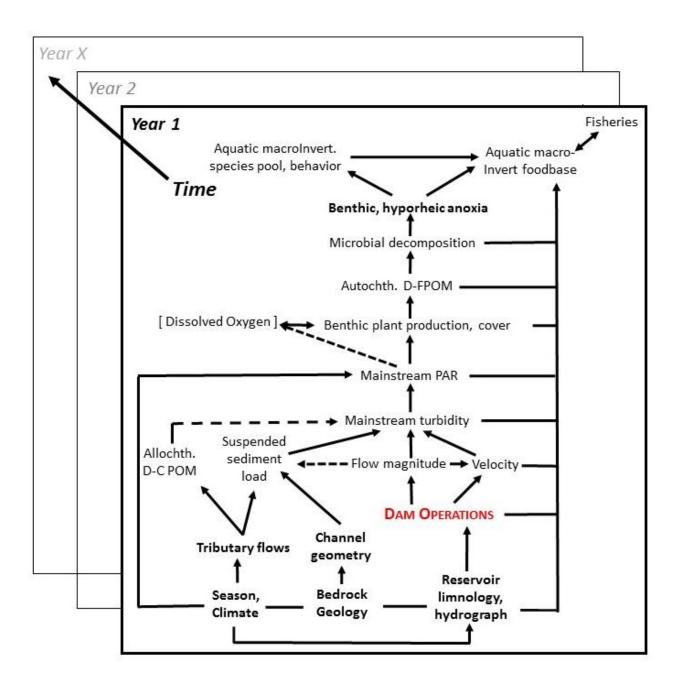


Fig. 20: Conceptual model of HA development through time. Bold text indicates stronger effects than non-bold text. Dashed lines indicate weaker effects than solid lines. "PAR" = photosynthetically active radiation. "DO" = dissolved oxygen. "D-C POM" = dissolved-to-coarse particulate organic matter. "DS" = downstream.

regulated river ecology has not been widely noted. Nearly 40 species of Chara have been described around the world, and several species such as C. vulgaris, are globally distributed. While nutrient uptake and mobilization from the benthos is considered to be relatively unimportant in most Characeae, several potential biogeochemical interactions may be of relevance in the CRE. Chara precipitates calcite during peak growth periods, a process that binds or sorbs P in calcite or benthic sediments. Chara may deliver dissolved O₂ into benthic substrata, enhancing nitrification, and preventing release of Fe-bound P from bed sediments. Physically, dense Chara beds can restrict groundwater flow (e.g., Banks et al. 2011), suspension of fine sediments, and temporally extended growth periods may increase Chara's influences on nutrient dynamics into the non-growing season. In addition, Chara may decompose more slowly than vascular aquatic taxa, extending the duration of Chara's nutrient influences in temperate latitudes. While Kufel and Kufel (2002) concluded that dense Chara stands function as nutrient traps in shallow lakes, Chara also has been widely reported to support N-fixing Cyanobacteria. Due to its ability to develop dense stands in lentic settings, it has been the subject of control efforts (e.g., Guha 1995). P is generally regarded to be a limiting nutrient in the Glen Canyon Dam tailwaters. If substantial N-fixation by Chara epiphytes is occurring in the Glen Canyon tailwaters, Chara may be influencing N : P ratios, development and persistence of BHA, and aquatic macroinvertebrate composition and structure.

The impacts of BHA on macroinvertebrates were mixed, in part because many of the subject organisms appear to be tolerant of BHA. Tolerant, non-EPT taxa, including dragonflies, damselfies, backswimmers, and non-native isopods and molluscs were little-affected by BHA solutes. Larval *Heptagenia* were used in the third, tightly controlled laboratory bioassay. This lotic mayfly species is abundant in coolwater tributary streams, but does not occur in the mainstream, and it is relatively intolerant of low water quality. Larval *Heptagenia* mortality in that experiment was significantly greater in the presence of BHA sediments than in the nosediment controls, possibly due to toxicity and/or physical obstruction of respiration by fine particulate organic BHA sediments. The only water bodies where mayfly and Odonata larvae occur in the study area are off-channel, groundwater-fed backwater habitats. This distribution pattern and the experiments support the hypothesis that BHA sediments may contribute to embeddedness and hinder colonization of potentially colonizing EPT populations in the mainstream. Additional studies are warranted to determine which of several flow, sediment, and geochemical mechanisms affect different potential macroinvertebrate colonizers (below).

A primary concern regarding BHA impacts on macroinvertebrates centers on the quality of the aquatic food base available to the fisheries of the Colorado River below Glen Canyon Dam. The macroinvertebrate community is depauperate in the mainstem of the Colorado River in Glen and Grand Canyons, and the specific mechanism(s) responsible for this are unclear. Kennedy et al. (2016) suggested that egg mortality due to frequent fluctuation in flows eliminates taxa that oviposit at the water's edge. Thus, egg mortality due to fluctuating flows may be the primary cause of low macroinvertebrate diversity and abundance in the mainstream. Our study suggests there may also be a negative population effect due to BHA development. With multiple mechanisms potentially hindering macroinvertebrate assemblage development, the aquatic food base for both native and non-native fish remains limited, and likely cannot develop into a robust condition without significant changes in river ecosystem management. Based on field mesocosm and laboratory microcosm experiments, the BHA development rate can be relatively rapid, occurring within 10 days, and potentially dissipating within 12 hr when exposed to the atmosphere. BHA development increased under conditions of warmer water temperatures, low dissolved oxygen concentration, and in the presence of *Chara* and possibly other aquatic macrophytes.

Potential Management Options

This preliminary study provides evidence for the mechanistic underpinnings of the development and maintenance of BHA in the Colorado River in Glen Canyon, and some potential ecosystem consequences of the phenomenon. Based on the results of our study and the subsequent conceptual model (Fig. 20), several separate but interrelated management actions could potentially decrease the extent and volume of BHA sediment in Glen Canyon.

(1) Increase mainstream DO concentration – Our laboratory microcosm experiment provides evidence that BHA development in sediment can be decreased via aeration. BHA is likely largely due to bacterial decomposition of aquatic vegetation, a process that consumes bed-sediment oxygen. Aeration of the low-oxygen hypolimnion water in Lake Powell may increase benthic dissolved oxygen downstream through several methods, including: deploying a bubbler or impeller in the forebay of Lake Powell reservoir or at the base of the dam, constructing turbines in the spillways (which could only be used when reservoir stage is near full-pool) or in the by-pass tubes, aerating water as it passes through the turbines, or by occasionally releasing water through the bypass tubes at Glen Canyon Dam. We demonstrated that BHA can develop within 10 days under constant lentic conditions (i.e., with Chara present, water temperatures of 20°C, and lack of aeration), and depths of BHA development can exceed 15 cm within a 38-day period. This rapid development rate suggests frequent or continually aerated water releases may be necessary to limit BHA development. Such actions appear to be most important during the growing season (mid-March through mid-October) when benthic macrophyte growth is greatest. This action would not eliminate BHA, and there are limitations to the success of this strategy; however, viewed in combination with (and as component of) the following two strategies, such actions can help ensure development of a benthic surficial oxygenated barrier sand layer. Of course, the frequency and duration of aerated releases will require further consideration and research.

(2) Increase mainstream turbidity – BHA develops in the presence of *Chara* and other benthic macrophytes at lower stage elevations in Lake Powell Reservoir and in the tailwaters, under well-lit and relatively stable (undisturbed) hydraulic conditions. However, turbidity (decreased benthic light availability) limits growth of submerged algae and macrophytic vegetation. Increased turbidity in the tailwaters reach has been explored as a management option, and while of interest in maintaining sandbars, its impacts on the trout fishery would likely be negative. Increased turbidity downstream from the mouth of the Paria River creates a stair-step, 30-fold reduction in benthic vegetation standing mass (Stevens et al. 1997) and appears to be the primary reason that BHA is only rarely detected downstream in Grand Canyon. Further turbidity addition may reduce food base availability for the downstream native and non-native fisheries. Periods of exceptionally stable, clearwater flows do occur below the Paria River, but apparently with insufficient frequency and/or duration to result in increased

benthic macrophytic cover. Therefore BHA development remains limited downstream in Grand Canyon.

(3) **Decrease mainstream temperature** – Cooler water holds more dissolved oxygen, and synergistically may slightly reduce BHA development. Cooler, more oxygenated water slightly reduces the extent of BHA development, and may be useful for maintaining the oxygenated buffer layer of benthic sand. Warming the mainstream temperature has been repeatedly suggested to improve native fish survivorship downstream; and both increased dissolved oxygen concentration and thermal control of the mainstream could be accomplished by placing turbines in the jet tubes. However, our experiments indicate that management for warmer water than presently released from Glen Canyon Dam could enhance the rate and extent of BHA development.

(4) **Increased flow variability** – BHA in the Glen Canyon Dam tailwaters reach developed after the onset of MLFF flows from Glen Canyon Dam in the mid-1990's, and has gradually come to dominate the channel bed there. Thus, reduced flow variability is likely at least partially responsible for BHA development. However, the timing and levels of flow variation needed to scour (reset) and retard or prevent subsequent BHA development requires further research. The 2016 HFE was not sufficient to strongly scour out much of the existing BHA, even though that high flow did promote development of the thin benthic buffer sand horizon. Rather than many small increases and decreases in water releases, fewer releases of greater magnitude followed by longer low-flow periods may have a larger negative impact on BHA development in the Glen Canyon tailwaters reach. Such a strategy could, during low flows, expose more of the affected sediments to the atmosphere, aerating the top layer of sediment, while simultaneously limiting the growth and survival of shallow submerged aquatic vegetation. If the opportunity occurs to release larger flows, one research theme should be to investigate the extent of BHA scour that occurs.

Another partial solution might be to physically disturb the benthic sediments prior to a high flow. Our bed-disturbance experiments demonstrated that even modest scour allows development of the benthic buffer sand horizon. A more robust bed disturbance experiment (e.g., power-washing) would help evaluate the cost : benefit ratio and timing trade-offs for that type of non-flow related management action. Timing such actions with HFEs might increase the longevity of effects, and we conclude there is much to be learned from such mechanically-simulated flood disturbance experiments.

CONCLUSIONS

Many uncertainties remain after this pilot examination of BHA in the Glen Canyon reach. Among them are the pending domination of the river bed by introduced quagga mussel (*Dreissena bugensis*). During the period of our study, we found quagga mussel density has already exceeded 1,000/m² on the floor of the river in the Glen Canyon reach. Bivalves are wellknown for their ability to filter water, removing sediment and phytoplankton and decreasing turbidity. The closely-related invasive zebra mussel (*D. polymorpha*) has been studied in the Great Lakes region for nearly three decades. Researchers have found that as mussel densities increase, turbidity subsequently decreases due to their powerful filtering action (Lowe and Pillsbury 1995, Skubinna et al. 1995, Churchill et al. 2016). In addition to increasing light availability via decreasing turbidity, dreissenid mussels also excrete additional nutrients into aquatic systems, some of which may affect aquatic production (i.e., P). This process may support additional growth of benthic filamentous algae (e.g., *Cladophora*) beyond what is expected in the absence of the invasive dreissenids (Francoeur et al. 2017). The mainstream channel floor was dominated by *Cladophora* prior to 1995 (Stevens et al. 1997), and recovery of Cladophora might reduce dominance of *Chara* and therefore reduce the extent of BHA. However, the impacts of quagga mussel invasion remain highly speculative at present.

While quagga mussel may remove nutrients, clarify the water, and set back benthic algal and macrophyte densities, the mussels are highly tolerant of BHA. Other non-native invertebrates also appear to be highly tolerant of BHA, including New Zealand mudsnails and *Gammarus lacustris* amphipods. Thus, as BHA developed in this highly regulated river tailwaters, it has indirectly supported expansion of a highly tolerant non-native aquatic macroinvertebrate assemblage.

The extent of BHA development documented in the Glen Canyon Dam tailwaters has affected virtually all of the Colorado River channel downslope of the 200 m³/sec stage elevation downstream from Glen Canyon Dam. Currently, the phenomenon appears to be limited to Lake Powell sand deposits and the tailwaters reach. BHA development is driven by increased water temperature, flow stability, and benthic plant growth, but it does not greatly alter the geochemistry of the water column. BHA appears to be detrimental to sensitive larval macroinvertebrates, like some mayfly species, possibly through physical and geochemical impacts on respiration. Consequently BHA may limit the potential aquatic macroinvertebrate assemblage and foodbase in the Glen Canyon reach. BHA does not presently occur to any great extent in the Grand Canyon reaches of the CRE. However, conditions that foster development of extensive *Chara* beds (i.e., reduced flow variability, decreased suspended sediment loads, warmer mainstream temperatures, and increased water clarity), may promote BHA development downstream.

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Appendix 1A:

Ephemeroptera, Plecoptera, and Trichoptera occurring in Grand Canyon (GC) or in the Grand Canyon ecoregion (GCE) detected (D), likely (L), or possibly (P) occurring, their EPA median tolerance score and functional feeding group (FFG; Merritt et al. 2008), elevation range (m), and range of flight dates.

		EPA Median		Elev Range	Flight Date		
Order	Full Name	Tolerance	FFG	(m)	Range	GCE	GC
EPH	Ameletidae Ameletus bellulus Zloty 1996	0	GC			Р	
EPH	Ameletidae Ameletus celer McDunnough 1934	0	GC		14 Jun-22 Aug	Р	
EPH	Ameletidae Ameletus cooki McDunnough 1929	0	GC			Р	
EPH	Ameletidae Ameletus doddsianus Zloty 1996	0	GC			L	
EPH	Ameletidae Ameletus sparsatus McDunnough 1931	0	GC	2120-2920		L	
EPH	Ameletidae Ameletus subnotatus Eaton 1885	0	GC			Р	
EPH	Ameletidae Ameletus velox Dodds 1923	0	GC	2680	10 Jul	D	
EPH	Ameletidae Ameletus vernalis McDunnough 1924	0	GC			Р	
EPH	Ametropodidae Ametropus neavei McDunnough 1928		GC			Р	
EPH	Baetidae Acentrella insignificans (McDunnough 1926)	4	GC	365-1770	8 Jun	D	D
EPH	Baetidae Acentrella turbida (McDunnough 1924)	4	GC			L	
EPH	Baetidae Acerpenna pygmaea (Hagen 1861)	4	SH			Р	
EPH	Baetidae Baetis adonis Traver 1935	5	GC	1220		L	D
EPH	Baetidae Baetis bicaudatus Dodds 1923	5	GC	1900-2920		L	
EPH	Baetidae Baetis brunneicolor McDunnough 1925	5	GC			Р	
EPH	Baetidae Baetis flavistriga McDunnough 1921	5	GC			Р	
EPH	Baetidae Baetis magnus McCafferty and Waltz 1986	5	GC	490-1900	14 Jan	D	D
EPH	Baetidae Baetis notos Allen and Murvosh 1987	5	GC	365-1770		D	D
EPH	Baetidae Baetis tricaudatus Dodds 1923	5	GC	590-2535	21 Jun-23 Jun	D	D
EPH	Baetidae Baetodes arizonensis Koss 1972			735-1220	18-Sep	D	D
EPH	Baetidae Baetodes deficiens Cohen and Allen 1972				•	Р	
EPH	Baetidae Baetodes edmundsi Koss 1972			600		D	D
EPH	Baetidae Callibaetis ferrugineus (Walsh 1862)	9	GC		2 Mar	Р	

Order	Full Name	EPA Median Tolerance	FFG	Elev Range (m)	Flight Date Range	GCE	GC
EPH	Baetidae Callibaetis ferrugineus hageni Eaton 1885	9	GC			D	
EPH	Baetidae Callibaetis fluctuans (Walsh 1862)	9	GC			Р	
EPH	Baetidae Callibaetis montanus Eaton 1885	9	GC			D	
EPH	Baetidae Callibaetis pallidus Banks 1900	9	GC			D	
EPH	Baetidae Callibaetis pictus (Eaton 1871)	9	GC	1463	4 May	D	
EPH	Baetidae Camelobaetidius			950		D	D
EPH	Baetidae Camelobaetidius kickapoo McCafferty 2000					L	
EPH	Baetidae Camelobaetidius warreni (Traver and Edmunds 1968)					L	
EPH	Baetidae Centroptilum album McDunnough 1926	2.7	GC			Р	
EPH	Baetidae Centroptilum asperatum Traver 1935	2.7	GC			L	
EPH	Baetidae Centroptilum conturbatum McDunnough 1929	2.7	GC			L	
EPH	Baetidae Cloeodes excogitatus Waltz and McCafferty 1987			560-790		D	D
EPH	Baetidae Diphetor hageni (Eaton 1885)	5	GC			L	
EPH	Baetidae Fallceon quilleri (Dodds 1923)		GC	365-2120	10 May-6 Nov	D	D
EPH	Baetidae Fallceon sonora (Allen and Murvosh 1987)		GC			Р	
EPH	Baetidae Moribaetis mimbresaurus McCafferty 2007			1710-1733	9 Feb-15 Mar	D	
EPH	Baetidae Paracloeodes minutus (Daggy 1945)	8.7	SC			Р	
EPH	Baetidae Pseudocloeon apache (McCafferty and Waltz 1995)	4	SC	1630-1770		D	
EPH	Baetidae Pseudocloeon dardanum (McDunnough 1923)	4	SC			Р	
EPH	Baetidae Pseudocloeon propinquum (Walsh 1863)	4	SC			Р	
EPH	Caenidae Caenis amica Hagen 1861	7	GC			L	
EPH	Caenidae Caenis bajaensis Allen and Murvosh 1983	7	GC			D	
EPH	Caenidae Caenis latipennis Banks 1907	7	GC			D	
EPH	Ephemerellidae Attenella delantala (Mayo 1952)	3	GC			Р	
EPH	Ephemerellidae Attenella margarita (Needham 1927)	3	GC			Р	
EPH	Ephemerellidae Drunella coloradensis (Dodds 1923)	0	PR	2680-2920	22 Aug	L	
EPH	Ephemerellidae Drunella doddsii (Needham 1927)	0	PR	2120-2820	-	L	
EPH	Ephemerellidae Drunella grandis (Eaton 1884)	0	PR	1770-2120		D	
EPH	Ephemerellidae Drunella grandis grandis (Eaton 1884)	0	PR			Р	
EPH	Ephemerellidae Drunella spinifera (Needham 1927)	0	PR			Р	
EPH	Ephemerellidae Ephemerella aurivillii (Bengtsson 1908)	2	GC			Р	
EPH	Ephemerellidae Ephemerella dorothea Needham 1908	2	GC			Р	

Order	Full Name	EPA Median Tolerance	FFG	Elev Range (m)	Flight Date Range	GCE	GC
EPH	Ephemerellidae Ephemerella excrucians Walsh 1862	2	GC	600-2500	13 Jan-8 Apr	D	L
EPH	Ephemerellidae Ephemerella tibialis McDunnough 1924	2	GC			L	
EPH	Ephemerellidae Serratella micheneri (Traver 1934)	2	GC			D	
EPH	Ephemerellidae Serratella tibialis McDunnough 1924	2	GC			L	
EPH	Ephemerellidae Timpanoga hecuba (Eaton 1884)	7	GC			Р	
EPH	Ephemeridae Ephemera simulans Walker 1853	2.7	GC			L	
EPH	Ephemeridae Hexagenia limbata (Serville 1829)	6	GC			Р	
EPH	Heptageniidae Cinygmula par (Eaton 1885)	4	SC		22 Aug	L	
EPH	Heptageniidae Ecdyonurus criddlei (McDunnough 1927)					D	
EPH	Heptageniidae Ecdyonurus simplicioides (McDunnough 1924)			2200	28 Jul	D	
EPH	Heptageniidae Epeorus albertae (McDunnough 1924)	1.2	SC			L	
EPH	Heptageniidae Epeorus deceptivus (McDunnough 1924)	1.2	SC		21 Sep	L	
EPH	Heptageniidae Epeorus grandis (McDunnough 1924)	1.2	SC			Р	
EPH	Heptageniidae Epeorus longimanus (Eaton 1885)	1.2	SC	605-2920	1 Jun-29 Jun	D	D
EPH	Heptageniidae Epeorus margarita Edmunds and Allen 1964	1.2	SC			D	
EPH	Heptageniidae Heptagenia adaequata McDunnough 1924	3	SC			L	
EPH	Heptageniidae Heptagenia elegantula (Eaton 1885)	3	SC	992		D	
EPH	Heptageniidae Heptagenia solitaria McDunnough 1924	3	SC	1630-1900		L	
EPH	Heptageniidae Leucrocuta petersi (Allen 1966)	1.7	SC			Р	
EPH	Heptageniidae Rhithrogena futilis McDunnough 1934	0.4	SC			Р	
EPH	Heptageniidae Rhithrogena hageni Eaton 1885	0.4	SC			L	
EPH	Heptageniidae Rhithrogena morrisoni (Banks 1924)	0.4	SC		19 May	D	
EPH	Heptageniidae Rhithrogena plana Allen and Chao 1978	0.4	SC			Р	
EPH	Heptageniidae Rhithrogena robusta Dodds 1923	0.4	SC	2680-2820		L	
EPH	Heptageniidae Rhithrogena undulata (Banks 1924)	0.4	SC		3 Jun	L	
EPH	Leptohyphidae Asioplax edmundsi (Allen 1967)					Р	
EPH	Leptohyphidae Homoleptohyphes dimorphus (Allen 1967)		1			D	
EPH	Leptohyphidae Homoleptohyphes quercus (Kilgore and Allen 1973)					D	
EPH	Leptohyphidae Leptohyphes lestes Allen and Brusca 1973	2				Р	
EPH	Leptohyphidae Leptohyphes zalope Traver 1958	2		480		D	Р
EPH	Leptohyphidae Tricoryhyphes condylus (Allen 1967)					D	

Onder	Euli Marca	EPA Median	FE0	Elev Range	Flight Date	005	
Order EPH	Full Name Leptohyphidae Tricorythodes explicatus (Eaton 1892)	Tolerance	FFG	(m) 900-1900	Range	GCE	
EPH	Leptohyphidae Tricorythodes minutus (Eaton 1892)			900-1900		D	Р
EPH	Leptophlebiidae Choroterpes inornata Eaton 1892	4	GC	975-1900	2 Apr	D	D
EPH	Leptophlebildae Neochoroterpes kossi (Allen 1974)	4	GC	975-1900	Z Api	D	
EPH	Leptophlebildae Neochoroterpes kossi (Allen 1974)					D	
EPH	Leptophlebiidae Paraleptophlebia bicornuta (McDunnough 1926)	1	GC			Р	
EPH	Leptophlebiidae Paraleptophlebia bicornuta (McDunnough 1926)	1	GC			Р	
EPH	Leptophlebiidae Paraleptophlebia debilis (Walker 1853)	1	GC	1630-2680	25 Sep	L	
EPH	Leptophlebiidae Paraleptophlebia heteronea (McDunnough 1924)	1	GC			Ρ	
EPH	Leptophlebiidae Paraleptophlebia memorialis (Eaton 1884)	1	GC	900-2120	3 Jun-4 Jun	D	D
EPH	Leptophlebiidae Thraulodes brunneus Koss 1966					Р	
EPH	Leptophlebiidae Thraulodes gonzalesi Traver and Edmunds 1967					L	
EPH	Leptophlebiidae Thraulodes speciosus Traver 1934					D	Р
EPH	Leptophlebiidae Traverella albertana (McDunnough 1931)			981		D	D
EPH	Oligoneuriidae Homoeoneuria alleni Pescador and Peters 1980				28 Sep	Р	
EPH	Oligoneuriidae Lachlania saskatchewanensis Ide 1941			991		D	
EPH	Siphlonuridae Siphlonurus columbianus McDunnough 1925	7	GC			Р	
EPH	Siphlonuridae Siphlonurus occidentalis (Eaton 1885)	7	GC	1463-2300	26 May-26 Jul	D	
PLE	Capniidae Bolshecapnia milami (Nebeker and Gaufin 1967)				18 Mar-1 Apr		
PLE	Capniidae Capnia arapahoe Nelson and Kondratieff 1988	1	SH	2012			
PLE	Capniidae Capnia coloradensis Claassen 1937	1	SH	2560			
PLE	Capniidae Capnia confusa Claassen 1936	1	SH			D	
PLE	Capniidae Capnia decepta (Banks 1897)	1	SH			D	
PLE	Capniidae Capnia gracilaria Claassen 1924	1	SH	2164-2560		D	
PLE	Capniidae Capnia limita	1	SH	2210-2560			
PLE	Capniidae Capnia nana Claassen 1924	1	SH				
PLE	Capniidae Capnia nelsoni Kondratieff and Baumann 2002	1	SH	3200			
PLE	Capniidae Capnia uintahi Gaufin 1964	1	SH				

		EPA Median		Elev Range	Flight Date		
Order	Full Name	Tolerance	FFG	(m)	Range	GCE	GC
PLE	Capniidae Capnia vernalis (Newport 1848)	1	SH	4500	20 Jan 20 Mar	D D	
PLE	Capniidae Capnura fibula (Claassen 1924)	1	SH	1500	30 Jan-28 Mar	P	
PLE	Capniidae Capnura wanica (Frison 1944)	1	SH	0000		P D	
PLE	Capniidae Eucapnopsis brevicauda Claassen 1924	1	SH	2622	40 Am 0 Max	D	
PLE	Capniidae Isocapnia crinita (Needham and Claassen 1925)	1	SH	2210	18 Apr-9 May		
PLE	Capniidae Isocapnia hyalita Ricker 1959	1	SH		6 May-5 Jul		
PLE	Capniidae Isocapnia vedderensis (Ricker 1943)	1	SH		1 May-3 Jun		
PLE	Capniidae Mesocapnia frisoni (Baumann and Gaufin 1970)	1	SH	930	5 Feb	D	
PLE	Capniidae Mesocapnia werneri (Baumann and Gaufin 1970)	1	SH			D	
PLE	Capniidae Paracapnia angulata Hanson 1961	1	SH				
PLE	Capniidae Utacapnia lemoniana (Nebeker and Gaufin 1965)	1	SH			D	
PLE	Capniidae Utacapnia logana (Nebeker and Gaufin 1965)	1	SH	2164-2560		D	
PLE	Capniidae Utacapnia poda (Nebeker and Gaufin 1965)	1	SH	2210-2560			
PLE	Chloroperlidae Alloperla pilosa Needham and Claassen 1925	1.2	PR				
PLE	Chloroperlidae Alloperla severa (Hagen 1861)	1.2	PR				
PLE	Chloroperlidae Paraperla frontalis (Banks 1902)	1	PR	2210-2911			
PLE	Chloroperlidae Plumiperla diversa (Frison 1935)	1	PR	2680-2920		D	
PLE	Chloroperlidae Suwallia lineosa (Banks 1918)	1	PR				
PLE	Chloroperlidae Suwallia pallidula (Banks 1904)	1	PR	2134-3170		D	Р
PLE	Chloroperlidae Suwallia starki Alexander and Stewart 1999	1	PR				
PLE	Chloroperlidae Suwallia wardi Kondratieff and Kirchner 1991	1	PR				
PLE	Chloroperlidae Sweltsa borealis (Banks 1895)	1	PR	2622-2941		D	
PLE	Chloroperlidae Sweltsa coloradensis (Banks 1898)	1	PR	2195-3170		D	
PLE	Chloroperlidae Sweltsa cristata Surdick	1	PR			Р	
PLE	Chloroperlidae Sweltsa fidelis (Banks 1920)	1	PR				
PLE	Chloroperlidae Sweltsa lamba (Needham and Claassen 1925)	1	PR	2100-2855	22 Jun-7 Sep	D	D
PLE	Chloroperlidae Triznaka pintada (Ricker 1952)	1	PR	2286		D	
PLE	Chloroperlidae Triznaka signata (Banks 1895)	1	PR	2195-2774		D	
PLE	Leuctridae Paraleuctra jewetti Nebeker and Gaufin 1966	0	SH				
PLE	Leuctridae Paraleuctra occidentalis (Banks 1907)	0	SH				
PLE	Leuctridae Paraleuctra projecta (Frison 1942)	0	SH			D	
PLE	Leuctridae Paraleuctra sara (Claassen 1937)	0	SH	2134-2804			

Order	Full Name	EPA Median Tolerance	FFG	Elev Range (m)	Flight Date Range	GCE	GC
PLE	Leuctridae Paraleuctra vershina Gaufin and Ricker 1974	0	SH	()	ittango		
PLE	Leuctridae Perlomyia utahensis Needham and Claassen 1925	0	SH	3084	24 Apr-14 Jun		
PLE	Nemouridae Amphinemura banksi		SH	2805	23 Aug	D	
PLE	Nemouridae Amphinemura banksi Baumann and Gaufin 1972	3	SH	2805	23 Aug	D	
PLE	Nemouridae Amphinemura mogollonica		SH	1930-1940	26 Oct	D	
PLE	Nemouridae Amphinemura mogollonica Baumann and Gaufin 1972	3	SH	1930	26 Oct	D	
PLE	Nemouridae Malenka californica (Claassen 1923)	2	SH				
PLE	Nemouridae Malenka coloradensis (Banks 1897)	2	SH	950-3155	4 Apr-15 Aug	D	D
PLE	Nemouridae Malenka flexura (Claassen 1923)	2	SH				
PLE	Nemouridae Podmosta decepta (Frison 1942)	2	SH				
PLE	Nemouridae Prostoia besametsa (Ricker 1952)	2	SH	2621-2880		D	
PLE	Nemouridae Zapada cinctipes (Banks 1897)	2	SH	2149-2560		D	
PLE	Nemouridae Zapada frigida (Claassen 1923)	2	SH			D	
PLE	Nemouridae Zapada haysi (Ricker 1952)	2	SH	2680-2920		D	
PLE	Nemouridae Zapada oregonensis (Claassen 1923)	2	SH	2865-2926			
PLE	Perlidae Acroneuria abnormis (Newman 1838)	0	PR			D	
PLE	Perlidae Anacroneuria wipukupa Baumann and Olson 1984		PR			D	
PLE	Perlidae Claassenia sabulosa (Banks 1900)	3	PR	2057-2713			
PLE	Perlidae Hesperoperla pacifica (Banks 1900)		PR	590-3170	22 Jun-29 Sep	D	D
PLE	Perlidae Isoperla longiseta Banks		PR			Р	
PLE	Perlidae Isoperla phalerata (Needham)		PR			Р	
PLE	Perlidae Neoperla clymene (Newman 1839)	1.6	PR				
PLE	Perlidae Perlesta decipiens (Walsh 1862)	4.8	PR				
PLE	Perlodidae Arcynopteryx compacta (McLachlan 1872)		PR	3560	16 Jul-8 Aug		
PLE	Perlodidae Cultus aestivalis (Needham and Claassen 1925)	2	PR	2103-2743			
PLE	Perlodidae Diura knowltoni (Frison 1937)	2	PR	2195-3170		D	
PLE	Perlodidae Isogenoides colubrinus (Hagen 1874)	2	PR				
PLE	Perlodidae Isogenoides elongatus (Hagen 1874)	2	PR	2134-2865			
PLE	Perlodidae Isogenoides zionensis Hanson 1949	2	PR	1610-2000	2 Jun	D	
PLE	Perlodidae Isoperla bilineata (Say 1823)	2	PR				
PLE	Perlodidae Isoperla fulva Claassen 1937	2	PR	2134-2804		D	

Order	Full Name	EPA Median Tolerance	FFG	Elev Range (m)	Flight Date Range	GCE	GC
PLE	Perlodidae Isoperla longiseta (Banks 1906)	2	PR			D	
PLE	Perlodidae Isoperla marlynia Needham and Claassen 1925	2	PR				
PLE	Perlodidae Isoperla mormona Banks 1920	2	PR	605-2286	23 Apr-8 Jun	D	D
PLE	Perlodidae Isoperla petersoni Needham and Christenson 1927	2	PR		12 Sep		
PLE	Perlodidae Isoperla phalerata (Smith 1917)	2	PR				
PLE	Perlodidae Isoperla pinta Frison 1937	2	PR	2195-2621		D	
PLE	Perlodidae Isoperla quinquepunctata (Banks 1902)	2	PR	605-2880	4 Apr-12 Jun	D	D
PLE	Perlodidae Isoperla sobria (Hagen 1874)	2	PR			D	
PLE	Perlodidae Kogotus modestus (Banks 1908)	2	PR	2682-2941			
PLE	Perlodidae Megarcys signata (Hagen 1874)	2	PR	2120-3170		D	
PLE	Perlodidae Pictetiella expansa (Banks 1920)	2	PR				
PLE	Perlodidae Skwala americana (Klapalek 1912)	2	PR	2103-2713			
PLE	Pteronarcyidae Pteronarcella badia (Hagen 1874)	0	SH	1630-3277		D	
PLE	Pteronarcyidae Pteronarcys californica Newport 1848	0	SH	2057-2377		D	
PLE	Taeniopterygidae Doddsia occidentalis (Banks 1900)	2	SC	3000			
PLE	Taeniopterygidae Oemopteryx fosketti (Ricker 1965)					Р	
PLE	Taeniopterygidae Taenionema pacificum (Banks 1900)	2	SC	2103-2667		D	
PLE	Taeniopterygidae Taenionema pallidum (Banks 1902)	2	SC	2210		D	
PLE	Taeniopterygidae Taenionema uinta Stanger and Baumann 1993	2	SC				
PLE	Taeniopterygidae Taeniopteryx burksi Ricker and Ross 1968	2	SH				
PLE	Taeniopterygidae Taeniopteryx parvula Banks 1918	2	SH			Р	Р
TRI	Apataniidae Apatania arizona Wiggins 1972	0.8	SC	730-2450	25 Apr-15 Aug	D	
TRI	Apataniidae Apatania comosa (Denning 1949)	0.8	SC		2 Jun	D	
TRI	Brachycentridae Brachycentrus americanus (Banks 1899)	1.6	FC	1606-3048	17 Apr-5 Oct	D	
TRI	Brachycentridae Brachycentrus echo (Ross 1947)	1.6	FC	1657-2073	7 Jul-11 Sep	D	
TRI	Brachycentridae Brachycentrus occidentalis Banks 1911	1.6	FC	1690-2926	3 Apr-25 Jul	D	D
TRI	Brachycentridae Micrasema bactro group Ross 1938	1.5	SH	605-1587	8-Jun	D	
TRI	Brachycentridae Micrasema bactro Ross 1938	1.5	SH	1100-2896	4 Oct-6 Mar	D	D
TRI	Brachycentridae Micrasema onisca Ross 1947	1.5	SH	730-2820	6 May-30 Sep	D	
TRI	Calamoceratidae Phylloicus aeneus (Hagen 1861)			1025-1550	25 Aug-28 Aug	D	

Order	Full Name	EPA Median Tolerance	FFG	Elev Range (m)	Flight Date Range	GCE	GC
TRI	Calamoceratidae Phylloicus mexicanus (Banks, 1900)				13 Mar-9 Sep	D	
TRI	Eunoidae Neophylax splendens Denning 1948			1920	12-27 Sep	L	
TRI	Glossosomatidae Agapetus boulderensis Milne 1936	0	SC	1370-2749	18 Jun-8 Sep	D	
TRI	Glossosomatidae Anagapetus debilis Ross 1938	0	SC		16-Aug	D	
TRI	Glossosomatidae Culoptila cantha (Ross, 1938)	0	SC	129-1801	8 Jan-17 Dec	D	
TRI	Glossosomatidae Culoptila kimminsi Denning 1965	0	SC			D	
TRI	Glossosomatidae Culoptila moselyi Denning 1965	0	SC		5 May-30 Jul	D	
TRI	Glossosomatidae Culoptila thoracica (Ross 1938)	0	SC	1690-3298	11 Jun-10 Aug	D	
TRI	Glossosomatidae Glossosoma ventrale Banks 1904	1.5	SC	1356-2691	13 Jan-13 Oct	D	
TRI	Glossosomatidae Glossosoma verdonum Ross 1938	1.5	SC	(1676) 1875-3094	(1 May) 20 May-18 Jul	D	
TRI	Glossosomatidae Protoptila balmorhea Ross, 1941	1	SC	1074		D	
TRI	Glossosomatidae Protoptila erotica Ross 1938	1	SC	959-1977	23 Apr-3 Aug	D	D
TRI	Helichopsychidae Helichopsyche borealis (Hagen 1861)	3	SC	500-3109	8 Jan-17 Dec	D	
TRI	Helichopsychidae Helichopsyche mexicana Banks 1901	3	SC	775-2550	11 Mar-19 Oct	D	D
TRI	Helichopsychidae Helicopsyche borealis (Hagen 1861)	3	SC	983-1820	22 Jun-3 Aug	D	
TRI	Hydrobiosidae Atopsyche sperryi Denning 1949	4		1346-2757	(24 May) 1 Jun-13 Aug	D	
TRI	Hydrobiosidae Atopsyche tripunctulata Banks 1905			1373-2541	42566	D	
TRI	Hydropsychidae Arctopsyche grandis (Banks 1900)	1	FC	950-3036	18 Mar-26 Sep	D	
TRI	Hydropsychidae Ceratopsyche cockerelli (Banks 1905)	2.7	FC	525-3109	10 May-8 Oct	D	D
TRI	Hydropsychidae Ceratopsyche oslari (Banks 1905)	2.7	FC	455-2895	27 Mar-13 Oct	D	
TRI	Hydropsychidae Ceratopsyche venada (Ross 1941)	2.7	FC	1025-2757	22 Apr-19 Jul (28 Aug)	D	D
TRI	Hydropsychidae Cheumatopsyche arizonensis (Ling 1938)	5	FC	959-2290	24 Feb-15 Oct	D	
TRI	Hydropsychidae Cheumatopsyche campyla Ross, 1938	5	FC	1690-1920	1-Jul	D	
TRI	Hydropsychidae Cheumatopsyche eonis Ross 1938	5	FC	1118-2333	19 Apr-23 Aug	D	
TRI	Hydropsychidae Cheumatopsyche gelita Denning 1952	5	FC	2114-2290	42540	D	
TRI	Hydropsychidae Cheumatopsyche lasia Ross 1938	5	FC	1555		Р	
TRI	Hydropsychidae Cheumatopsyche pinula Denning 1952	5	FC	1030-2525	28 Apr-4 Sep	D	D
TRI	Hydropsychidae Hydropsyche auricolor Ulmer 1905	4	FC	915-1707	5 Feb-4 Jul	D	D
TRI	Hydropsychidae Hydropsyche californca Banks 1899	4	FC	1200-1365	15 Jul-4 Sep	D	

		EPA Median		Elev Range	Flight Date		
Order	Full Name	Tolerance	FFG	(m)	Range	GCE	GC
TRI	Hydropsychidae Hydropsyche occidentalis Banks 1900	4	FC	565-2600 (2941)	20 Feb-30 Sep	D	
TRI	Hydropsychidae Hydropsyche quitchupaha Korecki and Ruiter 2009	4	FC		28 May (18 Jun)	D	
TRI	Hydropsychidae Parapsyche almota Ross 1938	1	PR	1981-2804	26 May - 7 Oct	D	
TRI	Hydropsychidae Potamyia sp.	2.5	FC			D	
TRI	Hydropsychidae Smicridea arizonensis Flint 1974			983-1757	22 Apr-4 Sep	D	
TRI	Hydropsychidae Smicridea dispar (Banks 1905), incl. S. utico Ross			983-1731	19 Apr-1 Sep (23 Dec)	D	
TRI	Hydropsychidae Smicridea fasciatella McLachlan 1871			45-1400	(19 Jan) 15 Feb-17 Dec	D	
TRI	Hydropsychidae Smicridea n.sp., Flint in prep.			950	17-Jun	D	
TRI	Hydropsychidae Smicridea signata (Banks 1903)			959-1805	17 May-23 Aug (25 Aug)	D	
TRI	Hydroptilidae Agraylea multipunctata Curtis 1834	6.9		1690-3500	30 Apr-24 Aug	D	
TRI	Hydroptilidae Alisotrichia arizonica (Blickle and Denning 1977)			1025-1250	30 Mar-30 Oct	D	D
TRI	Hydroptilidae Hydroptila ajax Ross 1938	6	SC	(70) 390- 1670	20 Jan-16 Dec	D	D
TRI	Hydroptilidae Hydroptila arctia Ross 1938	6	SC	400-2749	7 Feb-4 Oct (3 Dec)	D	
TRI	Hydroptilidae Hydroptila argosa Ross 1938	6	SC	1515-2633	3 May-1 Oct	D	Р
TRI	Hydroptilidae Hydroptila consimilis Morton 1905	6	SC	1576-2579	13 Feb-8 Oct	D	
TRI	Hydroptilidae Hydroptila hamata Morton 1905	6	SC	1010-2757	21-30 Jun	L	
TRI	Hydroptilidae Hydroptila icona Mosely 1937	6	SC	45-1820	9 May-21 Dec	D	
TRI	Hydroptilidae Hydroptila modica Mosely 1937	6	SC	940	8 Jun-13 Aug	D	
TRI	Hydroptilidae Hydroptila pecos Ross 1941	6	SC			Р	Р
TRI	Hydroptilidae Hydroptila rono Ross 1941	6	SC	1525-3048	3 Apr-25 Jul	D	
TRI	Hydroptilidae Ithytrichia clavata Morton, 1905			1821		D	
TRI	Hydroptilidae Ithytrichia mexicana Harris and Contreras-Ramos 1989			2290-2600	9 Jun-25 Jul (30 Jul)	D	D
TRI	Hydroptilidae Leucotrichia limpia Ross 1944	6	SC	390-1550	5 Aug-18 Dec	D	
TRI	Hydroptilidae Leucotrichia pictipes (Banks 1911)	6	SC	1384-2120	20 Feb-30 Oct	D	

		EPA Median		Elev Range	Flight Date		
Order	Full Name	Tolerance	FFG	(m)	Range	GCE	GC
TRI	Hydroptilidae Leucotrichia sarita Ross 1944	6	SC			D	
TRI	Hydroptilidae Mayatrichia acuna Ross 1944	6	SC	975-1850	12 May-11 Aug	D	
TRI	Hydroptilidae Mayatrichia ayama Mosely 1937	6	SC	959-1030	11 Jul-10 Oct	D	D
TRI	Hydroptilidae Mayatrichia ponta Ross 1944	6	SC	390	22-Aug	D	
TRI	Hydroptilidae Metrichia arizonensis (Flint 1972)			1020		D	
TRI	Hydroptilidae Metrichia nigritta (Banks 1907)			983-1375	13-May	D	
TRI	Hydroptilidae Neotrichia blinni Ruiter	3.6	SC	2600	1-Jul	D	
TRI	Hydroptilidae Neotrichia caxima Mosely 1937	3.6	SC	450	19 Aug-10 Oct	D	
TRI	Hydroptilidae Neotrichia downsi Ruiter 1990	3.6	SC	1585-1890	6 May-23 Aug	L	
TRI	Hydroptilidae Neotrichia halia Denning 1948	3.6	SC	1010-2490	22-Aug	Р	
TRI	Hydroptilidae Neotrichia okopa Ross 1939	3.6	SC	1401-1525	1 May-10 Aug	D	
TRI	Hydroptilidae Neotrichia olorina (Mosely 1937)	3.6	SC	129-240	42590	D	
TRI	Hydroptilidae Neotrichia osmena Ross 1944	3.6	SC	1646-1700	27 Jun-16 Aug	D	
TRI	Hydroptilidae Neotrichia sonora Ross 1944	3.6	SC	1010-1621	10-Aug	D	
TRI	Hydroptilidae Ochrotrichia argentea Flint and Blickle 1972	4	GC			D	D
TRI	Hydroptilidae Ochrotrichia arizonica Denning and Blickle 1972	4	GC	1200	15 Jun-28 Jul	D	
TRI	Hydroptilidae Ochrotrichia dactylophora Flint 1965	4	GC	650-2743	11 May-2 Jul	D	D
TRI	Hydroptilidae Ochrotrichia ildria Denning and Blickle, 1972	4	GC	1050-2596	22 Jun-14 Aug	D	D
TRI	Hydroptilidae Ochrotrichia logana (Ross 1941)	4	GC	475-1737	15 Jun-4 Aug	D	D
TRI	Hydroptilidae Ochrotrichia lometa (Ross 1941)	4	GC	736-2600	19 Jun-23 Sep	D	
TRI	Hydroptilidae Ochrotrichia quadrispina Denning and Blickle 1972	4	GC	1403-1999	5 May-1 Jun	D	
TRI	Hydroptilidae Ochrotrichia rothi Denning and Blickle 1972	4	GC			Р	D
TRI	Hydroptilidae Ochrotrichia stylata (Ross 1938)	4	GC	535-2579	27 May-19 Jun	D	
TRI	Hydroptilidae Ochrotrichia tarsalis (Hagen 1861)	4	GC	959-1850	16-30 Jun	D	D
TRI	Hydroptilidae Orthotrichia cristata Morton 1905	6	SC	375	13-Nov	R	
TRI	Hydroptilidae Oxyethira aculea Ross 1941	5.2		573-2757		D	
TRI	Hydroptilidae Oxyethira arizona Ross 1948	5.2		129-647	28 Mar-12 Jul	D	
TRI	Hydroptilidae Oxyethira dualis Morton 1905	5.2		573-3098	9 May-23 Aug (23 Nov)	L	
TRI	Hydroptilidae Stactobiella brustia (Ross 1938)	2	SH	1519-2579	25-28 Jul	Р	

Order	Full Name	EPA Median Tolerance	FFG	Elev Range (m)	Flight Date Range	GCE	GC
TRI	Hydroptilidae Zumatrichia notosa (Ross 1944)			1000-1707	11 May-26 Jul	D	
TRI	Hydroptilidae Zumatrichia notosa (Ross 1944)			950-1707	11 May-4 Sep	D	
TRI	Lepidostomatidae Lepidostoma apornum Denning 1949	1	SH	1618-2644	4 May-22 Jun	D	
TRI	Lepidostomatidae Lepidostoma bakeri Flint 1965	1	SH	1690-1865	2 Apr-8 Aug	L	
TRI	Lepidostomatidae Lepidostoma knulli Ross 1946	1	SH	372-2596	18-Jun	D	
TRI	Lepidostomatidae Lepidostoma mexicanum (Banks 1901)	1	SH			L	
TRI	Lepidostomatidae Lepidostoma ormea Ross 1946	1	SH	1690-2530	(1 Jun) 18 Jun-22 Aug	D	D
TRI	Lepidostomatidae Lepidostoma pluviale (Milne 1936)	1	SH	1020-2749	9 Jun-30 Sep	D	
TRI	Lepidostomatidae Lepidostoma roafi (Milne 1936)	1	SH	1926-3109	22 Jun-29 Sep	D	
TRI	Leptoceridae Ceraclea annulicornis (Stephens 1836)	3	GC	2420	24-Jul	Р	Р
TRI	Leptoceridae Nectopsyche albida (Walker 1852)	3	SH	250-1052	29 May-11 Jul	D	
TRI	Leptoceridae Nectopsyche dorsalis (Banks 1901)	3	SH	129-1755	20 Apr-1 Oct	D	L
TRI	Leptoceridae Nectopsyche gracilis (incl. intervena) (Banks 1901)	3	SH	455-1707	25-28 Jul	D	
TRI	Leptoceridae Nectopsyche lahontensis Haddock 1977	3	SH	1053-2384	23-Apr	Р	Р
TRI	Leptoceridae Nectopsyche minuta (Banks 1900)	3	SH	1400-1800	(20 Jul)	Р	
TRI	Leptoceridae Nectopsyche spilosa (Banks 1901)	5	SH	115	Ca 15 Apr-15 Nov		
TRI	Leptoceridae Nectopsyche stigmatica (Banks 1901)	3	SH	983-1829	23 Apr-6 Aug	D	
TRI	Leptoceridae Oecetis arizonica Denning 1951	8	PR	1375-2556	13 May-25 Jul	D	D
TRI	Leptoceridae Oecetis avara (Banks 1895)	8	PR	410-2490	19 Apr-15 Oct	D	Р
TRI	Leptoceridae Oecetis disjuncta (Banks 1920)	8	PR	940-2797	8 Jun-13 Aug	D	Р
TRI	Leptoceridae Oecetis inconspicua (Walker 1852)	8	PR	45-2896	9 May-8 Aug	D	
TRI	Leptoceridae Oecetis metlacensis Bueno-Soria 1981	8	PR	2334		D	
TRI	Leptoceridae Triaenodes (Ylodes) reuteri McLachlan 1880	6		(890) 1690- 2920	(12 May) 19 Jun-17 Jul	D	
TRI	Leptoceridae Triaenodes (Ylodes) frontalis Banks 1907	6		1690-2805	15 Jul-4 Sep	D	
TRI	Leptoceridae Triaenodes tardus Milne 1934	6				L	
TRI	Limnephilidae Allomyia gnathos (Ross 1950)	0	SC	2713-3616	19 Jun	L	
TRI	Limnephilidae Amphicosmoecus canax (Ross 1947)		SH	1765-2911	8 Oct-11 Jan	D	
TRI	Limnephilidae Anabolia bimaculata (Walker 1852)		SH	1690-3200	12 Jun-28 Aug	D	

		EPA Median		Elev Range	Flight Date		
Order	Full Name	Tolerance	FFG	(m)	Range	GCE	GC
TRI	Limnephilidae Asynarchus nigriculus (Banks 1908)			2621-3687	~15 Jun-12 Aug	Р	
TRI	Limnephilidae Chyrandra centralis (Banks 1900)	1	SH	2680-3658	18 Jun-15 Aug	D	
TRI	Limnephilidae Clistoronia formosa (Banks 1900)					Р	
TRI	Limnephilidae Clistoronia maculata (Banks 1904)			2074-2797	20 Jun-23 Jul	D	
TRI	Limnephilidae Crenophylax (Limnephilus) sperryi (Banks 1914)			2590-2765	20 Jun-5 Aug	D	
TRI	Limnephilidae Dicosmoecus atripes (Hagen 1875)	1	SH	1690-3688	18 Jul-21 Aug	D	
TRI	Limnephilidae Ecclisomyia maculosa (Schmid 1955; E. simulata Banks 1920)	2	GC	2558-3377	25 May-5 Aug	D	
TRI	Limnephilidae Glyphopsyche irrorata (Fabricius 1781)	1		2999	9-Apr	Р	
TRI	Limnephilidae Hesperophylax consimilis (Banks 1900)	5	SH	1690-2743	13 Feb-12 Sep	D	
TRI	Limnephilidae Hesperophylax d. designatus (Walker 1852)	5	SH	1690-2425	11-Aug	D	D
TRI	Limnephilidae Hesperophylax magnus Banks 1918	5	SH	720-2757	3 Apr-29 Sep	D	
TRI	Limnephilidae Hesperophylax occidentalis (Banks 1908)	5	SH	1220-3667	2 Apr-1 Oct	D	
TRI	Limnephilidae Limnephilus acnestus Ross 1938	5	SH			Р	
TRI	Limnephilidae Limnephilus apache Flint 1965	5	SH	2130-2530	22 Jun-14 Aug	D	D
TRI	Limnephilidae Limnephilus arizona Ross 1941	5	SH	1850-2520	26 Jun-21 Aug	D	
TRI	Limnephilidae Limnephilus assimilis (Banks 1908)	5	SH	(685)-697	(30 Apr)-15 Oct	D	
TRI	Limnephilidae Limnephilus bucketti Denning 1965	5	SH	1070	23-Sep	D	
TRI	Limnephilidae Limnephilus diversus (Banks 1903)	5	SH	1440-2530	26 Jul-13 Aug	D	
TRI	Limnephilidae Limnephilus frijole Ross 1944	5	SH	1265-2271	22 Jun-8 Oct	D	
TRI	Limnephilidae Limnephilus granti Nimmo 1991	5	SH	2495-2596	8-Jun	D	
TRI	Limnephilidae Limnephilus lithus (Milne 1935)	5	SH	1500-2600	9 Jun-15 Oct	D	
TRI	Limnephilidae Limnephilus moestus Banks 1908	5	SH	2749	22-Jul	D	
TRI	Limnephilidae Limnephilus productus Banks 1914	5	SH	1597-2896	3-29 Aug	D	
TRI	Limnephilidae Limnephilus secludens Banks 1914	5	SH			Р	
TRI	Limnephilidae Limnephilus spinatus Banks 1914	5	SH	1829	30 May-3 Sep	D	D
TRI	Limnephilidae Limnephilus tulatus Denning 1962	5	SH	560-1800	25 Jun-26 Oct	D	
TRI	Limnephilidae Onocosmoecus unicolor (Banks 1897)	1	SH	1690-3124	19 Jul-30 Aug	D	
TRI	Limnephilidae Psychoglypha schuhi Denning 1970	1	GC	2539-3616	1-Jul	D	Р
TRI	Limnephilidae Psychoglypha subborealis (Banks 1924)	1	GC	1690-2757	1 May-6 Oct	D	D

		EPA Median		Elev Range	Flight Date		
Order	Full Name	Tolerance	FFG	(m)	Range	GCE	GC
TRI	Odontoceridae Marilia flexuosa Ulmer 1905			365-2507	28 May-27 Sep	D	
TRI	Odontoceridae Marilia nobsca Milne 1936			983-1773	22 Apr-25 Jul	D	
TRI	Philopotamidae Chimarra angustipennis Banks 1903	4	FC	560-1555	11 Jun-16 Dec	D	
TRI	Philopotamidae Chimarra elia Ross 1944	4	FC	560	28 Mar-17 Dec	D	
TRI	Philopotamidae Chimarra primula Denning 1950	4	FC	1373-1731	26 Jun-26 Aug	D	D
TRI	Philopotamidae Chimarra ridleyi Denning 1941	4	FC	360-1707	2 Apr-2 Oct	D	D
TRI	Philopotamidae Chimarra utahensis (=idahoensis) Ross 1938	4	FC	475-2507	30 Oct-20 Feb	D	
TRI	Philopotamidae Dolophilodes novusamericana (Ling 1938)	1	GC	1697-2650	(6 Feb) 25 May-12 Aug (11 Oct)	D	D
TRI	Philopotamidae Wormaldia arizonensis (Ling 1938)	1.7	FC	790-1681	27 Nov-29 Feb	D	
TRI	Philopotamidae Wormaldia planae Ross and King 1956	1.7	FC	1078-1280	19 May-30 Aug	D	
TRI	Phryganeidae Agrypnia deflata (Milne 1931)			2736-3181	20 Jun-22 Aug	D	
TRI	Polycentropodidae Polycentropus arizonensis Banks 1905	5	PR	1373-2600	14 Jan-2 Sep	D	
TRI	Polycentropodidae Polycentropus aztecus Flint 1966	5	PR	2490-2600	23-Jul	D	
TRI	Polycentropodidae Polycentropus flavus (Banks 1908)	5	PR	1643	4-Aug	D	
TRI	Polycentropodidae Polycentropus gertschi Denning 1950	5	PR	1123-2530	20 May-4 Sep	D	D
TRI	Polycentropodidae Polycentropus halidus Milne 1936	5	PR	412-1829	8 May-15 Oct	D	
TRI	Polycentropodidae Polycentropus variegatus Banks 1900	5	PR	1000-2682	15 Apr-30 Sep	L	
TRI	Psychomyiidae Psychomyia flavida Hagen 1861	2	SC	1444-2920	6 Jun-4 Sep	D	D
TRI	Psychomyiidae Tinodes provo Ross and Merkley 1950	2	SC	(565) 750- 525	29 Feb-26 Sep	D	
TRI	Rhyacophilidae Rhyacophila acropedes Banks 1914 (Smith and Manuel 1984)	0	PR			D	
TRI	Rhyacophilidae Rhyacophila alberta Banks 1918	0	PR	2536-3295	4-22 Aug	D	
TRI	Rhyacophilidae Rhyacophila angelita Banks 1911	0	PR	1975-3036	7 Jul-9 Oct	D	
TRI	Rhyacophilidae Rhyacophila brunnea Banks 1911	0	PR	1690-3261	2 Apr-16 Sep	D	D
TRI	Rhyacophilidae Rhyacophila c. coloradensis Banks 1904	0	PR	3319	26 Mar-25 Oct	D	
TRI	Rhyacophilidae Rhyacophila chordata Denning 1989	0	PR	~2000	(27 Feb) 28 May-16 Jun	D	
TRI	Rhyacophilidae Rhyacophila harmstoni Ross 1944	0	PR	2000-3475	18 Apr-15 Aug	D	

Order	Full Name	EPA Median Tolerance	FFG	Elev Range (m)	Flight Date Range	GCE	GC
TRI	Rhyacophilidae Rhyacophila hyalinata Banks 1905	0	PR	2264-3542	20 Jul-15 Sep	D	
TRI	Rhyacophilidae Rhyacophila jenniferae Peck, 1978	0	PR	1829	(1 May) 6 May-29 Jun	L	D
TRI	Rhyacophilidae Rhyacophila kernada Ross 1950	0	PR	880	5-Apr	D	
TRI	Rhyacophilidae Rhyacophila pellisa Ross 1938	0	PR	2286-3194	8 May-21 Aug	D	D
TRI	Rhyacophilidae Rhyacophila rotunda Banks 1924	0	PR	690-2950	2 Apr-2 Aug	D	D
TRI	Sericostomatidae Gumaga griseola (McLachlan 1871)	3	SH	1190-2797	15 Jun-3 Aug	D	D
TRI	Sericostomatidae Gumaga nigricula (McLachlan, 1871)	3	SH	890-2130	>1 Jun	D	
TRI	Uenoidae Neothremma alicia Dodds and Hisaw 1925	0	SC	1900-3170	26 May-28 Sep (10 Oct)	D	D
TRI	Uenoidae Oligophlebodes minutus Banks (1897)	1	SC	1020-3094	(5 May) 21 Jun-15 Sep (30 Sep)	D	
TRI	Uenoidae Oligophlebodes sierra Ross 1944	1	SC	2853	26-Jul	D	L
TRI	Uenoidae Oligophlebodes sigma Milne 1935	1	SC	2541-2757	>29 Jun - 7 Jul	D	
TRI	Xiphocentronidae Cnodocentron yavapai Moulton and Stewart 1997			1000	23 Apr-10 May	D	

Appendix 1B:

Water quality range data for Lees Ferry (first row in *bold-italic* font, in post-dam time) and for Ephemeroptera, Plecoptera, and Trichoptera genera occurring in Grand Canyon or in the Grand Canyon ecoregion, where data were available.

Order	Family	Genus	pH Range	% DO Saturation Range	DO Range (mg/L)	Total Alkalinity Range (mg/L)	Water Temp Range (°C)	Specific Conductance Range (µS/cm)
Lees Ferry			6.44 - 8.40		5.67- 11.30	116-154	6.9-21.2	509-851
EPH	Ameletidae	Ameletus	-	-	-	-	-	-
EPH	Ametropodidae	Ametropus	-	-	-	-	-	-
EPH	Baetidae	Acentrella	-	-	-	-	-	-
EPH	Baetidae	Baetis	7.6-8.7	95-95	7.25-7.25	-	5.8-14.4	286-572
EPH	Baetidae	Baetodes	7.97- 7.97	95-95 67.4-	7.25-7.25	-	-	432-432
EPH	Baetidae	Callibaetis	6.2-8.8	122.3	4.5-9.6	-	7-31.4	27-4531
EPH	Baetidae	Centroptilum	-	-	-	-	-	-
EPH	Baetidae	Cloeodes	-	-	-	-	-	-
EPH	Baetidae	Diphetor	-	-	-	-	-	-
EPH	Baetidae	Fallceon	7.97- 8.23	95-95	7.25- 11.47	-	23.4-23.4	432-491
EPH	Baetidae	Heterocloeon	-	-	-	-	-	-
EPH	Baetidae	Moribaetis	-	-	-	-	-	-
EPH	Baetidae	Paracloeodes	-	-	-	-	-	-
EPH	Baetidae	Procloeon	-	-	-	-	-	-

Order	Family	Genus	pH Range	% DO Saturation Range	DO Range (mg/L)	Total Alkalinity Range (mg/L)	Water Temp Range (°C)	Specific Conductance Range (µS/cm)
EPH	Baetidae	Pseudocloeon	-	-	-	-	-	-
EPH	Caenidae	Caenis	-	-	-	-	-	-
EPH	Ephemerellidae	Attenella	-	-	-	-	-	-
EPH	Ephemerellidae	Caudatella	-	-	-	-	-	-
EPH	Ephemerellidae	Drunella	-	-	-	-	-	-
EPH	Ephemerellidae	Ephemerella	-	-	-	-	-	-
EPH	Ephemerellidae	Serratella	-	-	-	-	-	-
EPH	Ephemerellidae	Timpanoga	-	-	-	-	-	-
EPH	Ephemeridae	Ephemera	-	-	-	-	-	-
EPH	Ephemeridae	Hexagenia	-	-	-	-	-	-
EPH	Heptageniidae	Cinygmula	-	-	-	-	-	-
EPH	Heptageniidae	Epeorus	7.6-8.4	-	-	-	5.8-8.8	323-354
EPH	Heptageniidae	Heptagenia	-	-	-	-	-	-
EPH	Heptageniidae	Leucrocuta	-	-	-	-	-	-
EPH	Heptageniidae	Rhithrogena	-	-	-	-	-	-
EPH	Heptageniidae	Stenacron	-	-	-	-	-	-
EPH	Isonychiidae	Isonychia	-	-	-	-	-	-
EPH	Leptohyphidae	Leptohyphes	-	-	-	-	-	-
EPH	Leptohyphidae	Tricorythodes	6.9- 8.23	83.1-95	5.16- 11.47	83.1-83.1	15.8-24.7	432-1047
EPH	Leptophlebiidae	Choroterpes	-	-	-	-	-	-
EPH	Leptophlebiidae	Leptophlebia	-	-	-	_	-	-
		· ·	8.23-		11.47-			101 101
EPH	Leptophlebiidae	Paraleptophlebia	8.23 7.97-	-	11.47 7.25-	-	23.4-23.4	491-491
EPH	Leptophlebiidae	Thraulodes	7.97- 8.23	95-95	7.25- 11.47	-	23.4-23.4	432-491
EPH	Leptophlebiidae	Traverella	-	-	-	-	-	-
EPH	Oligoneuriidae	Lachlania	-	-	-	-	-	-

Order	Family	Genus	pH Range	% DO Saturation Range	DO Range (mg/L)	Total Alkalinity Range (mg/L)	Water Temp Range (°C)	Specific Conductance Range (µS/cm)
EPH	Polymitarcyidae	Ephoron	-	-	-	-	-	-
EPH	Siphlonuridae	Siphlonurus	-	-	-	-	-	-
PLE	Capniidae	Capnia	-	-	-	-	-	-
PLE	Capniidae	Capnura	-	-	-	-	-	-
PLE	Capniidae	Eucapnopsis	-	-	-	-	-	-
PLE	Capniidae	Isocapnia	-	-	-	-	-	-
PLE	Capniidae	Mesocapnia	-	-	-	-	-	-
PLE	Capniidae	Paracapnia	-	-	-	-	-	-
PLE	Capniidae	Utacapnia	-	-	-	-	-	-
PLE	Chloroperlidae	Alloperla	-	-	-	-	-	-
PLE	Chloroperlidae	Paraperla	-	-	-	-	-	-
PLE	Chloroperlidae	Plumiperla	-	-	-	-	-	-
PLE	Chloroperlidae	Suwallia	8.78- 8.78	-	-	251.25- 251.25	9.7-9.7	396-396
PLE	Chloroperlidae	Sweltsa	7.7-8.7	-	-	225-246.25	5.8-22.1	135-380
PLE	Chloroperlidae	Triznaka	-	-	-	-	-	-
PLE	Leuctridae	Paraleuctra	-	-	-	-	-	-
PLE	Leuctridae	Perlomyia	-	-	-	-	-	-
PLE	Nemouridae	Amphinemura	-	-	-	-	-	-
PLE	Nemouridae	Malenka	6.56-9	-	5.93- 6.614	21.25-318	5.7-22.1	135-604
PLE	Nemouridae	Podmosta	-	-	-	-	-	-
PLE	Nemouridae	Prostoia	-	-	-	-	-	-
PLE	Nemouridae	Zapada	7.58- 7.58	-	7.713- 7.713	254-254	11.81- 11.81	307.15- 307.15
PLE	Perlidae	Acroneuria	-	-	-	-	-	-
PLE	Perlidae	Anacroneuria	-	-	-	-	-	-
PLE	Perlidae	Claassenia	-	-	-	-	-	-

Order	Family	Genus	pH Range	% DO Saturation Range	DO Range (mg/L)	Total Alkalinity Range (mg/L)	Water Temp Range (°C)	Specific Conductance Range (µS/cm)
PLE	Perlidae	Hesperoperla	-	-	-	-	-	-
PLE	Perlidae	Neoperla	-	-	-	-	-	-
PLE	Perlidae	Perlesta	-	-	-	-	-	-
PLE	Perlodidae	Arcynopteryx	-	-	-	-	-	-
PLE	Perlodidae	Cultus	-	-	-	-	-	-
PLE	Perlodidae	Diura	-	-	-	-	-	-
PLE	Perlodidae	Isogenoides	7.58- 7.58 7.58-	-	7.713- 7.713 7.713-	254-254	11.81- 11.81	307.15- 307.15
PLE	Perlodidae	Isoperla	7.56-	-	7.713	254-254	8.6-13.5	307.15-354
PLE	Perlodidae	Kogotus	-	-	-	-	-	-
PLE	Perlodidae	Megarcys	-	-	-	-	-	-
PLE	Perlodidae	Pictetiella	-	-	-	-	-	-
PLE	Perlodidae	Skwala	-	-	-	-	-	-
PLE	Pteronarcyidae	Pteronarcella	-	-	-	-	-	-
PLE	Pteronarcyidae	Pteronarcys	-	-	-	-	-	-
PLE	Taeniopterygidae	Doddsia	-	-	-	-	-	-
PLE	Taeniopterygidae	Oemopteryx	-	-	-	-	-	-
PLE	Taeniopterygidae	Taenionema	-	-	-	-	-	-
PLE	Taeniopterygidae	Taeniopteryx	-	-	-	-	-	-
PLE	Taeniopterygidae		-	-	-	-	-	-
TRI	Apataniidae	Allomyia	-	-	-	-	-	-
TRI	Apataniidae	Apatania	7.55- 7.6	-	-	23.75-23.75	8.3-11.7	361-479
TRI	Brachycentridae	Amiocentrus	-	-	-	-	-	-
TRI	Brachycentridae	Brachycentrus	-	-	-	-	-	-
TRI	Brachycentridae	Micrasema	-	-	-	-	-	-
TRI	Calamoceratidae	Phylloicus	-	-	-	-	-	-

Order	Family	Genus	pH Range	% DO Saturation Range	DO Range (mg/L)	Total Alkalinity Range (mg/L)	Water Temp Range (°C)	Specific Conductance Range (µS/cm)
TRI	Glossosomatidae	Agapetus	-	-	-	-	-	-
TRI	Glossosomatidae	Anagapetus	-	-	-	-	-	-
TRI	Glossosomatidae	Culoptila	-	-	-	-	-	-
TRI	Glossosomatidae	Glossosoma	7.6-7.7	-	-	-	8.6-8.8	347-354
TRI	Glossosomatidae	Protoptila	-	-	-	-	-	-
TRI	Helicopsychidae	Helicopsyche	6.9-8.2	83.1- 83.1	5.34-5.34	-	13.5-24.7	567-920
TRI	Hydrobiosidae	Atopsyche	-	-	-	-	-	-
TRI	Hydropsychidae	Arctopsyche	-	-	-	-	-	-
TRI	Hydropsychidae	Ceratopsyche	7.5-7.5	-	-	-	13.5-16.3	646-646
TRI	Hydropsychidae	Cheumatopsyche	7.24- 8.23	66.4- 66.4	4.65- 11.47	-	22.3-23.9	474-665
TRI	Hydropsychidae	Hydropsyche	8-8.2	-	-	-	14.4-19.6	332-395
TRI	Hydropsychidae	Helicopsyche	-	-	-	-	-	-
TRI	Hydropsychidae	Parapsyche	-	-	-	-	-	-
TRI	Hydropsychidae	Potamyia	-	-	-	-	-	-
TRI	Hydropsychidae	Smicridea	-	-	-	-	-	-
TRI	Hydroptilidae	Agraylea	-	-	-	-	-	-
TRI	Hydroptilidae	Alisotrichia	-	-	-	-	-	-
TRI	Hydroptilidae	Ithytrichia	-	-	-	-	-	-
TRI	Hydroptilidae	Ithytrichia	-	-	-	-	-	-
TRI	Hydroptilidae	Hydroptila	8.2-8.2	-	-	-	13.5-13.5	920-920
TRI	Hydroptilidae	Leucotrichia	-	-	-	-	-	-
TRI	Hydroptilidae	Zumatrichia	-	-	-	-	-	-
TRI	Hydroptilidae	Mayatrichia	-	-	-	-	-	-
TRI	Hydroptilidae	Metrichia	-	-	-	-	-	-
TRI	Hydroptilidae	Neotrichia	-	-	-	-	-	-
TRI	Hydroptilidae	Ochrotrichia	7.7-7.7	-	-	-	8.8-13.5	354-354

Order	Family	Genus	pH Range	% DO Saturation Range	DO Range (mg/L)	Total Alkalinity Range (mg/L)	Water Temp Range (°C)	Specific Conductance Range (µS/cm)
TRI	Hydroptilidae	Orthotrichia	-	-	-	-	-	-
TRI	Hydroptilidae	Oxyethira	-	-	-	-	-	-
TRI	Hydroptilidae	Stactobiella	-	-	-	-	-	-
TRI	Lepidostomatidae	Lepidostoma	7.6-7.7	-	-	-	8.6-8.8	347-354
TRI	Leptoceridae	Ceraclea	-	-	-	-	-	-
TRI	Leptoceridae	Nectopsyche	-	-	-	-	13.5-13.5	-
TRI	Leptoceridae	Oecetis	8.01- 8.01	-	7.67-7.67	-	-	474-474
TRI	Leptoceridae	Ylodes	-	-	-	-	-	-
TRI	Leptoceridae	Triaenodes	-	-	-	-	-	-
TRI	Limnephilidae	Chyrandra	-	-	-	-	-	-
TRI	Limnephilidae	Allomyia	-	-	-	-	-	-
TRI	Limnephilidae	Amphicosmoecus	-	-	-	-	-	-
TRI	Limnephilidae	Anabolia	-	-	-	-	-	-
TRI	Limnephilidae	Asynarchus	-	-	-	-	-	-
TRI	Limnephilidae	Clistoronia	-	-	-	-	-	-
TRI	Limnephilidae	Crenophylax	-	-	-	-	-	-
TRI	Limnephilidae	Dicosmoecus	-	-	-	-	-	-
TRI	Limnephilidae	Ecclisomyia	-	-	-	-	-	-
TRI	Limnephilidae	Glyphopsyche	-	-	-	-	-	-
TRI	Limnephilidae	Grammotaulius	-	-	-	-	-	-
TRI	Limnephilidae	Hesperophylax	6.2-9	-	6.614- 6.614	36.25-290	5.2-16.7	27-763
TRI	Limnephilidae	Homophylax	-	-	-	-	-	-
TRI	Limnephilidae	Lenarchus	-	-	-	-	-	-
TRI	Limnephilidae	Limnephilus	6.6-9.2	-	-	-	5.4-21	63-646
TRI	Limnephilidae	Nemotaulius	-	-	-	-	-	-
TRI	Limnephilidae	Neophylax	-	-	-	-	-	-

Order	Family	Genus	pH Range	% DO Saturation Range	DO Range (mg/L)	Total Alkalinity Range (mg/L)	Water Temp Range (°C)	Specific Conductance Range (µS/cm)
TRI	Limnephilidae	Oligophlebodes	-	-	-	-	-	-
TRI	Limnephilidae	Onocosmoecus	-	-	-	-	-	-
TRI	Limnephilidae	Philacrtus	-	-	-	-	-	-
TRI	Limnephilidae	Psychoglypha	-	-	-	-	-	-
TRI	Limnephilidae	Psychoronia	-	-	-	-	-	-
TRI	Molanidae	Molanna	-	-	-	-	-	-
TRI	Odontoceridae	Marilia	6.9- 8.23	83.1- 83.1	5.34- 11.47	-	20.3-24.7	474-679
TRI	Odontoceridae	Parthina	-	-	-	-	-	-
TRI	Philopotamidae	Chimarra	6.9- 8.41	83.1- 83.1	5.34-7.67	-	13.5-24.7	474-1446
TRI	Philopotamidae	Dolophilodes	-	-	-	-	-	-
TRI	Philopotamidae	Wormaldia	6.62- 8.23	-	5.49- 11.47	-	13.5-23.4	418-646
TRI	Phryganeidae	Agrypnia	-	-	-	-	-	-
TRI	Polycentropodidae	Plectrocnemia	-	-	-	-	-	-
TRI	Polycentropodidae	Polycentropus	7.2- 7.39	-	6.07-6.07	-	14.5-23.1	582-582
TRI	Psychomyiidae	Psychomyia	-	-	-	-	-	-
TRI	Psychomyiidae	Tinodes	-	-	-	-	13.5-13.5	-
TRI	Rhyacophilidae	Rhyacophila	7.6-9	-	-	-	5.7-8.8	286-354
TRI	Sericostomatidae	Gumaga	6.98- 8.2	-	5.16-8.23	-	12.1-20.3	485-1047
TRI	Uenoidae	Neothremma	-	-	-	-	-	-
TRI	Uenoidae	Oligophlebodes	8.4-8.5	-	-	-	5.8-5.8	322-323
TRI	Xiphocentronidae	Cnodocentron	-	-	-	-	-	-

Appendix 2:

Project field and laboratory data (Submitted electronically in Microsoft Excel format)