ANNUAL REPORT

MACROINVERTEBRATE INVESTIGATION: CHELAN RIVER, WA

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Prepared by

Terraqua, Inc.

for the

Public Utility District #1 of Chelan County

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Project Location

Chelan River, Chelan County, Washington T27N R22E 13; R23E 18-19, 29-30

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ABSTRACT

This annual report summarizes data collection and analysis results for the 2016 implementation of the Chelan River Macroinvertebrate Investigation (Chelan PUD 15-73). The results of this study are intended to provide a baseline for measuring success in meeting the Biological Objectives outlined in the Lake Chelan Settlement Agreement. We describe the biomass and taxonomic diversity of drift and benthic macroinvertebrate communities encountered in the restored sections of the Chelan River in May and August of 2016, and compare these communities with upstream and downstream reference sites and comparison streams in the same ecoregion. We found that the biological diversity of the benthic community in the restored sections of the Chelan River was generally comparable or greater than in the reference sections, but lower in taxa richness (B-IBI = 14-24) than regional comparison streams. The drift community was similar in biomass and diversity to regional comparison streams. Fieldwork and data analysis were completed by Terraqua, Inc., and laboratory and analytical support were provided by Rhithron Associates, Inc. for the Public Utility District #1 of Chelan County.

INTRODUCTION

This study took place in the Chelan River, which drains Lake Chelan, WA (T27N R22E 13; R23E 18-19, 29-30; Figure 1) into the upper Columbia River. The Lake Chelan Hydroelectric Project (Dam; FERC No. 637) serves a dual purpose of generating power and regulating the level of Lake Chelan. The Lake Chelan Settlement Agreement (SA; October 2003) was developed during the FERC relicensing process for the Project. The SA established a minimum flow of 80 cfs for the Chelan River, which had previously been dry from August-May in most years since the dam began operation in 1926, and called for habitat improvement features in an engineered "habitat channel" and the dam's tailrace to provide spawning habitat in the lower river. A number of criteria were established by the SA to measure components leading to success in achieving the Biological Objectives, including water quality requirements and standards for egg to fry survival. Other monitoring and evaluation activities specified in the SA include fish surveys and monitoring of benthic macroinvertebrate populations, which is the subject of this research project.

Mandatory monitoring and evaluation activities that have been implemented through the SA track and document progress towards achievement of established Biological Objectives and provide information to inform adaptive management strategies. The Biological Objective in Reaches 1-3 of the Chelan River is to create habitat to support a viable cutthroat trout population of 200 fish. The Biological Objectives for the Habitat Channel and tailrace spawning areas are to provide spawning and rearing habitat for Chinook salmon and steelhead, to document that these fish are using the new habitat, and to show evidence that adult fish production (returning adults) originated from fish spawning in this habitat.

The macroinvertebrate population structure in the Chelan River is previously unstudied, except for samples in the tailrace in 1999. Macroinvertebrate colonization of the upper river is probably limited to aerial colonization or downstream drift of invertebrates or material via spillway input from Lake Chelan, which may be dominated by taxa not suited to residence in riverine habitat. The tailrace and Habitat Channel may be populated through all three possible routes: aerial colonization, downstream drift, and upstream dispersal. Productivity in all reaches of the Chelan River may be limited by high summer stream temperatures, poor nutrient input from the highly oligotrophic Lake Chelan, and subject to periods of possible scouring during regulated spill. Any macroinvertebrate population prior to initiation of minimum flows in October 2009 was likely eradicated seasonally when the river went dry.

The goal of this project was to determine baseline condition of the benthic and drift macroinvertebrate population assemblage in the Chelan River in order to provide a metric for measuring success in meeting the Biological Objectives outlined in the Lake Chelan SA. The study area encompassed the entire Chelan River excluding the gorge, which is considered poor habitat for macroinvertebrates and unsafe for researcher access. The river was stratified into four primary areas of interest: 1) above the Lake Chelan Dam (0.75 rkm); 2) "Reach 1" (sub-stratified into upper, middle and lower sections) between the dam and the top of the Chelan River Gorge (3.45 rkm); 3) the engineered Habitat Channel located within "Reach 4" (0.55 rkm); and 4) the powerhouse tailrace near the Columbia River confluence (0.2 rkm) (Figure 1). In the SA, the area above the Lake Chelan Dam and the powerhouse tailrace were specified as

reference areas, while the restoration objective for Reach 1 and the Habitat Channel was to achieve a benthic community with comparable or greater density and species diversity than these reference areas.

Water in Reach 1 flows from Lake Chelan either through a low-level outlet structure or from the spillway. The bed of this relatively low gradient (1%) section is primarily composed of large cobbles and small boulders, with smaller cobbles and gravels generally limited to the margins of the river channel. This reach is moderately confined by hill slopes composed of glacial moraine deposits. Most fine bed materials are flushed out of the river during annual spill events, but pockets of medium-sized cobble and small gravels exist in some areas. Channel width through Reach 1 averages 28 m, and is primarily confined to a single channel except for a short (~640 m) braided section near the lower end of the reach. Riparian vegetation is scarce throughout Reach 1, with the most significant stands of riparian cover existing along the braided section.

The Habitat Channel is an engineered sinuous stream channel parallel to and upstream of the main tailrace. It is watered by the mainstem Chelan River, but has supplemental flow pumped from the tailrace during peak salmonid spawning periods in the spring and fall. Substrate varies from large cobbles to small gravel and some areas of sands. Riparian vegetation is thick, and primarily dominated by willows.

The section of the Chelan River above the dam is backwatered and typically slow water velocity and depths >2 m. Substrate is composed of small and large cobbles, gravels, sand and some fines. The section of river below the tailrace and Habitat Channel contains similar substrate and depths, but is also influenced by eddy flows as it joins the Columbia River. It is primarily watered by the tailrace but also includes flows from the Habitat Channel and an ephemeral floodplain channel, primarily hugging the north shore of this section.

The project collected drift and benthic macroinvertebrate samples. Concurrent metrics included stream flow velocity and water temperature at each drift net transect, water temperature at each benthic sample site, and alkalinity within each stratum at the time of sampling. The study leveraged existing Chelan PUD and U.S. Geological Survey data sources for average annual temperature and total stream discharge.

Objectives

- Assess abundance, taxonomic classification and biological health of the benthic macroinvertebrate community of the Chelan River;
- Assess biomass, abundance, taxonomic classification, resource category and size distribution of the drift macroinvertebrate community of the Chelan River;
- Assess biomass of organic debris in the Chelan River;
- Identify taxonomic classification of the benthic macroinvertebrate community immediately upstream of the Lake Chelan Hydroelectric Dam and the benthic and drift macroinvertebrate communities immediately downstream of the tailrace in order to determine the contribution of these habitats via upstream dispersal or downstream drift to the macroinvertebrate communities in the Chelan River; and

• Compare Chelan River macroinvertebrate community structure with that of comparable stream systems, with an emphasis, where possible, on other lake-fed, warm-water salmonid-bearing streams in the Pacific Northwest.

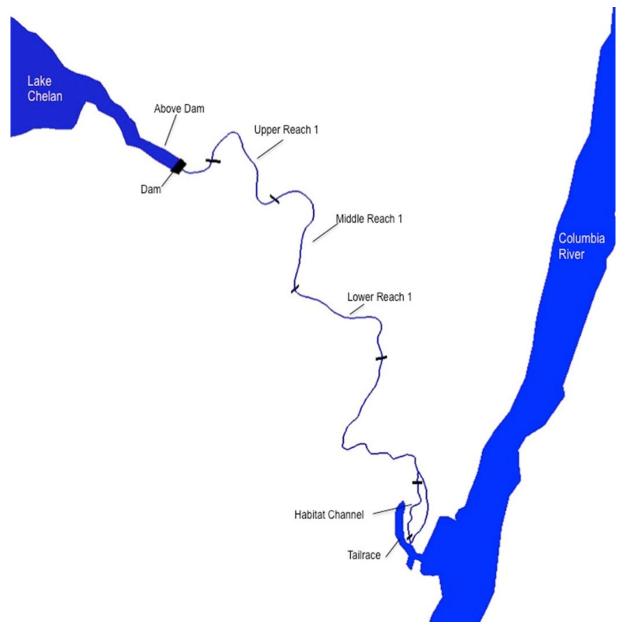


FIGURE 1. Study area within the Chelan River, WA in 2016. Benthic samples were collected in all 6 strata. Drift samples were collected in all strata except Above Dam.

METHODS

Organization and Schedule

Sampling was scheduled to represent the macroinvertebrate community during two seasons. Spring sampling took place May 2-4, 2016, and summer/fall sampling took place August 30-31, 2016. An additional benthic sample was collected above the dam in July to improve data quality as discussed later (Figure 2). Sampling in both seasons was scheduled to avoid conflict with concurrent fish snorkel surveys being conducted in multiple reaches, and to allow safe and effective sampling below an average 85 cfs discharge threshold. This was challenging during the May sample event when discharge had to be increased at the spillway to mitigate for an unseasonably high lake surface elevation, and the sampling timeframe was, in essence, contrived. Discharge averaged 86 cfs throughout the sampling timeframe in August (Figure 3), and was irrelevant in July as sampling only took place above the dam. Access permission was obtained from the Chelan PUD, and sites were accessed by vehicle and on foot, except for above the dam where a small motorized vessel was used.

Above Dam	Benthic						
Reach 1 Upper	Drift						
Reach i Opper	Benthic						
Reach 1 Middle	Drift						
Reach i Midule	Benthic						
Reach 1 Lower	Drift						
Reach i Lower	Benthic						
Habitat Channel	Drift						
	Benthic						
Tailrace	Drift						
Tulliuoc	Benthic						
	2-	May 3 [.]	May	4-May	14-Jul	30-Aug	31-Aug

FIGURE 2. Sampling schedule for drift and benthic macroinvertebrate sampling in the Chelan River, 2016.

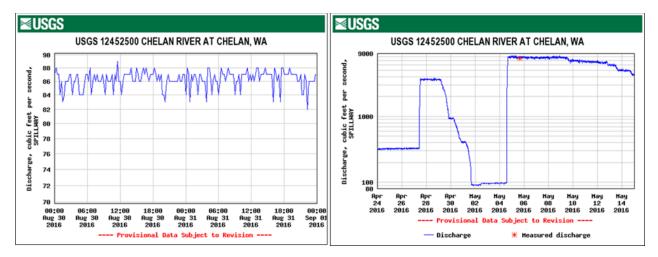


FIGURE 3. Chelan River discharge (cfs) measured ¹/₄ mile downstream from Chelan Dam (from waterdata.usgs.gov), showing ramp down window for sampling May 2-4, 2016 and measured discharge during sampling Aug. 30-31, 2016.

Sampling Process Design

Drift Sampling

We used a probabilistic design for drift macroinvertebrate site selection within Reach 1 and the Habitat Channel, and a targeted design in the tailrace section, with a total of five drift samples collected during each of the sample events (May and August). No drift samples were collected during the extra July sample event as this was targeted exclusively at collecting a replacement benthic sample above the dam. A master sample list defined potential drift sampling transects at 50 m intervals along the linear extent of each sampling area. Transects were then assigned a random number (rank) using MS Excel, and sorted by rank to assign use order within each strata. If the first use order site had to be rejected for any reason, then the next use order site was chosen for sampling. Rejected transects are permanently replaced (Appendix A). Reach 1 was divided into three spatially balanced strata (upper, middle, lower), and the Habitat Channel comprised a single stratum. Drift nets were set in suitable habitat, per protocol, within a maximum of 50 m upstream of each selected transect. The same drift transects were sampled during the spring and summer/fall sample events in Reach 1 and the Habitat Channel. A targeted site was chosen for drift sampling in the Tailrace, with a different site chosen for each season because of modified channel conditions. Each site represented the best available substrate for protocol adherence at the time of the event.

Benthic Sampling

Benthic macroinvertebrate sampling sites were selected randomly by field crews to represent eight different riffle or fast-water habitats within each stratum, using professional judgment to determine suitable sites during each season. All of the eight sites within each of the six strata (Upper, Middle and Lower Reach 1, Habitat Channel, Above-Dam and Tailrace) were composited into a single sample representing that stratum, for a total of six benthic samples collected during the May and August sample events, and one benthic sample during the July sample event.

Additional Data

Stream flow, depth and temperature were measured at the inlet of the drift sampling nets at each transect at the beginning and end of each set. Stream temperature and depth were also measured concurrent to each replicate benthic sample collected. Alkalinity was measured near the mid-point of each stratum, once per sample event. GPS coordinates were recorded for all replicates. Spillway data were provided in real time by Chelan PUD and summary discharge data were provided by USGS.

Sampling Procedures

Benthic sampling within wadeable areas (all but Above Dam) followed Washington Department of Ecology (WA-DOE) and Pacific Northwest Aquatic Monitoring Program (PNAMP) protocols (Adams 2010, Hayslip 2007). Within each of the five wadeable sampling strata, a total of 8 ft² of stream bottom was sampled and composited into a single sample for taxonomic processing, as previously discussed. One sample jar was used for each stratum, but samples could be split into multiple jars if additional capacity were needed. All wadeable benthic samples were collected using a 1 ft x 1 ft D-frame kick net with 500 μ m mesh. Some samples collected within the tailrace stratum exceeded recommended depth for sampling, making collection difficult but possible. All samples were deemed viable and representative of the population in this area. Water depth, temperature and GPS coordinates were recorded at each replicate site.

In May, a modified benthic sampling protocol was employed above the dam, using an extendable D-frame kick net and brush. However, poor sample quality forced revision of this method, and in July we tested dredge sampling as an alternative method. Thereafter, benthic samples were collected by boat in the stratum above the Dam, using a 6"x6"x6" AMS Ekman dredge sampler. A total of 8 replicate samples were collected, filtered through a 500 μ m mesh net and composited into a single sample for taxonomic processing. All samples were deemed viable and representative of the population in this area.

Drift sampling followed Bonneville Power Administration's Columbia Habitat Monitoring Program protocols (CHaMP 2015). At each of the five transects, two replicate samples were collected within suitable riffle or fast-water habitat. Drift nets (40 cm x 20 cm, 1000µm mesh) were set for a minimum of 3 hours at each transect. Replicate nets were set as far apart as possible for the available habitat. All drift samples were collected starting at least 2 hours after sunrise, and completed at least 2 hours before sunset. Replicate samples were considered as a single sample per transect for taxonomic processing, and were contained in one sample jar per stratum. Water temperature, depth and flow velocity entering the mouth of each net was recorded at the start and end of sampling. GPS coordinates were also collected for each net location.

Containers, Preservation, Holding Times

All samples containing invertebrates and/or organic matter were retained in plastic sample jars with 95% ethanol. Jars were labeled with project name, site ID or stratum, date, time, replicate and sample type. All sample jars were stored on ice or in a refrigerator within 8 hours of collection, and shipped (decanted prior to shipping) to Rhithron for taxonomic processing within 24 hours of sampling conclusion. Chain of Custody (CoC) forms were included in each shipment. Samples were received by Rhithron in good condition for each sample event, and recharged with ethanol for storage and subsequent taxonomic processing.

Data Management and Quality Assurance/ Quality Control

Field metadata were recorded electronically using a custom data collection form built in MDC GISCloud on an Apple iPad. Metadata associated with the start and end of each replicate drift sample included site ID, date, time, stream temperature, depth, flow velocity and GPS coordinates. Metadata associated with each benthic sample included stratum, replicate number, date, time, stream temperature and GPS coordinates. Additionally, alkalinity measurements with associated GPS coordinates and date/time were recorded for each stratum on the date of sampling. The entire survey extent was within range of cellular data services, and therefore each data entry was automatically and immediately backed up to a cloud server. Data were downloaded and backed up to a laptop computer at the end of each day of sampling, and consolidated into a single MS Excel database file at the completion of all sampling. Data QA/QC was completed by the lead investigator within 4 weeks following each sampling event. No errors or omissions exist in the data, all physical samples were present and accounted for, and all metadata are deemed to be complete and accurate. Taxonomic processing was completed and data delivered by Rhithron on August 9th (May, July) and October 3rd (August).

Taxonomic Analysis

Benthic samples were sorted according to Plotnikoff and Wiseman (2001) and drift samples were sorted according to CHaMP protocols to obtain representative subsamples with a minimum of 500 or 600 organisms, respectively. Briefly, samples were mixed before sorting and evenly spread onto a Caton sub-sampling tray (Caton 1991) with 30 grids. The contents of randomly selected grids were sorted and individual taxa were identified until the minimum number of organisms was reached. After obtaining the subsample, benthic samples were then scanned for large or rare individuals. Total body length was measured for drift samples. If the individual was damaged, body length was estimated by comparing with other individuals of the same taxon and maturity stage.

After obtaining the subsamples, drift samples were coarsely sorted by resource class (aquatic, terrestrial, aquatic/terrestrial) for dry biomass measurement. Filters (47mm glass, Whitman Glass-Fiber Filters types GF/A, 1.6 micron pore size) in aluminum boats were preashed at 500°C for 20 minutes, placed in a desiccation chamber to cool to room temperature, and weighed. Filters were placed in a filtration apparatus and moistened with de-ionized water before samples were added. Filters were dried at 105°C until constant weight was reached for a minimum of 2 hours and weighed. The same method was used to measure ash free dry weight of detritus.

Quality control was performed to assess initial sample processing and subsampling. On a random selection of 10% of the samples an independent sorter re-examined 25% of the sorted substrate using the same Caton grid method described above. Organisms that were missed were counted and added to the results from the original sort. Taxonomic identification was checked by an independent taxonomist by randomly selecting two samples and re-identifying all organisms. Sorting efficiency was 99.1% and taxonomic precision was 97.9%. Both are within industry standards (Stribling *et al.* 2003).

Data Analysis

Multivariate analyses were used to explore differences at the assemblage level and assess the similarities of taxonomic diversity and biomass among strata, as used by Favaro *et al.* (2014) and Maitland *et al.* (2016), for benthic and drift communities separately. Among the many similarity indices used for ordination in community ecology research, the Bray-Curtis similarity index (Clarke and Warwick 2001) has been found to be one of the best methods (see Quinn and Keough 2002). In this study, Bray-Curtis similarity indices (Clarke and Warwick 2001) were calculated for all strata pairs based on macroinvertebrate abundance data, and ordinated using non-metric multidimensional scaling (NMDS). NMDS is unconstrained by environmental variables, and thus reflects only similarities between taxonomic composition data.

For the benthic data, six sample strata (Above Dam, Reach 1 Upper, Reach 1 Middle, Reach 1 Lower, Habitat Channel, Tailrace) were compared, and for the drift data the same strata were compared except for the Above Dam strata which was not sampled for drift, making a total of five sample strata for drift data. We combined the species abundance data of both sampling periods to perform the NMDS because of the small sample sizes and high variations between the two sampling periods.

Goodness of fit (stress value) was also calculated in the ordination analysis for both communities (benthic and drift), in order to determine how well the ordination summarizes the observed distances between strata. For this study, we could not determine the relationship between community composition and environmental conditions because of limited environmental data so we illustrated how much the communities varied between strata.

Once we determined the variation of community assemblages, we further calculated similarities between sample strata for each sampling period, and a combination of both between paired strata in order to understand the contribution of organisms between strata via upstream or downstream dispersal. We hypothesized that downstream dispersal of organisms from above the dam would result in community structures of high similarity between the three main reaches (Above Dam, Reach 1, Tailrace/Habitat Channel), and that significantly different community structure between these reaches meant that downstream dispersal was not a significant mechanism for recruitment.

For this hypothesis, we used both the Bray-Curtis similarities index (Clarke and Warwick 2001) and Sørensen similarities index (Chao *et al.* 2005). Bray-Curtis similarities index (Clarke

and Warwick 2001) is based on taxonomic abundance data; however, in some cases taxa abundance and its variation or noise is often high, which might create a bias in the results (Pandit *et al.* 2009). We therefore also used Sørensen similarities index (Chao *et al.* 2005), which is based only on taxa presence/absence; and also provides greater weight to taxa common to all strata than to those found in only one stratum. The following equations were used for this study for both benthic and drift community structure.

Bray-Curtis Similarities Index:

$$d^{BCD}(i,j) = 1 - \frac{\sum_{k=0}^{n-1} |y_{i,k} - y_{j,k}|}{\sum_{k=0}^{n-1} (y_{i,k} + y_{j,k})}$$

where d^{BCD} is the similarity in community compositions between two strata, $y_{i,k}$ is the taxa at stratum one, and y_{jk} is the taxa at stratum two.

Sørensen Similarities Index:

$$1 - \frac{2a}{(2a+b+c)}$$

where a is the number of species common to both strata, b is the number of species unique to the first stratum, and c is the number of species unique to the second stratum.

Benthic Index of Biotic Integrity (B-IBI) were calculated by Rhithron for each strata/period using standardized equations (Fore and Wisseman 2012; Karr and Chu 1999). Ten individual metric scores ranging from one through 10 are added together to generate a total B-IBI score, which ranges from 1-100. B-IBI scores are used as an indicator of overall stream health based on undisturbed reference streams, with ranges representing: Very Poor [0,20), Poor [20,40), Fair [40,60), Good [60,80), or Excellent [80,100]. These qualifiers are part a standard scoring system, independent of the stream or ecoregion, and are commonly used to compare B-IBI scores based on other reference streams in the area. However, they do not necessary mean that a stream is of poor health just because it has a low score. For example, if a site has a low B-IBI score but surrounding reference sites also have low B-IBI scores, the site is geographically isolated from recruitment sources for indicator taxa, or the sampled area contains mostly habitat naturally unsuited to benthic colonization, we could conclude that this score represents the natural condition of the stream, independent of disturbance (Elbrecht *et al.* 2014).

We compared B-IBI metric scores for Chelan River benthic macroinvertebrate communities to other streams in Eastern Washington to estimate the status of colonization and diversity for baseline conditions. Comparison categories included reference streams, lake-fed streams, high-temperature streams, and the Palouse River, which best matched the Chelan River for annual flow regime and average summer temperatures. All data used for comparison were the most recent available and for which a majority of sampling occurred in July-August summer conditions; Chelan River data were collected primarily in spring and late summer/fall conditions. Benthic reference streams are included in the WA-DOE's Ambient Stream Biological Monitoring Project (ASBMP). The streams included in this project were chosen as a point of comparison for what benthic invertebrate compositions might be expected given pristine or undisturbed conditions. Reference streams are categorized into eight Washington ecoregions, which are considered to be areas of similar benthic invertebrate compositions (Wiseman 2003) within a similar range of environmental conditions. We chose nine reference streams (see Appendix B) that were geographically closest to the Chelan River with elevations under 600 m and bankfull widths greater than 30 m. However, given the Chelan River's unique geomorphology as compared to any of the reference streams existing in the ASBMP database, the nine reference streams chosen do not necessarily represent what pristine conditions might look like in the Chelan watershed, or even represent an attainable restoration objective for this system, but were the best available data.

We could not source data for any lake-fed streams with similar discharge and temperature as the Chelan River within our ecoregion, so we compared lake-fed steams and high temperature streams separately. Data were available for two lake-fed rivers in Eastern Washington with benthic invertebrate data: 1) the Cle Elum River and 2) the confluence of the Yakima and Cle Elum Rivers below where the Cle Elum River flows out of Lake Cle Elum. The Cle Elum River flows from Hyas Lake in the Alpine Lakes Wilderness area, the site is colder (10.5°C) and at a higher elevation (1,054 m) than the Chelan River, and although discharge data is not available it is likely lower. The site at the confluence of the Yakima and Cle Elum Rivers has higher discharge (3,700 cfs) but similar high average summer temperature (18.7°C) to the Chelan River, although this is still much cooler than the >23°C summer peak water temperatures that enter the Chelan River from the outlet of Lake Chelan.

We found three Eastern Washington streams with similar temperature ranges as the Chelan River (19.2 - 23°C). Discharge data was not available for these streams, so we used our best judgment to exclude rivers with much higher or lower discharge than the Chelan River. The Palouse River, a tributary to the Snake River, was chosen as the best match to the Chelan River for both temperature and discharge regimes in the Eastern Washington ecoregion. It has temperatures in the mid-20s in August, similar summer low flows, and large intra-annual discharge variations.

A standardized biotic index scoring system has not been developed to use for drift macroinvertebrate communities as the ASBMP has done for benthic macroinvertebrate communities in Washington. However, a common use of drift macroinvertebrate data is to compare total biomass with bioenergetic needs of fish in order to predict carrying capacity. We compared drift macroinvertebrate taxonomic diversity and total biomass for the Chelan River to other rivers within the CHaMP network. Additionally, we investigated similarities in community composition by taxa presence/absence at all comparison streams using Sørenson Similarities Indices (Chao *et al.* 2005).

CHaMP collects drift macroinvertebrate data at hundreds of sites annually throughout the Columbia River Basin. We chose to narrow down comparison stream selection to the Wenatchee, Methow and Entiat River subbasins, which were the closest geographic proximity to the Chelan River. Data were not available for any directly lake-fed rivers in the CHaMP network, so comparison streams were chosen based on similar channel width and discharge

profiles to the Chelan River. Temperature profiles were not directly compared, but all comparison streams experience high temperatures (>18°C) at times during the summer. None of the comparison streams can be considered as directly analogous to the Chelan River, again due to the unique geomorphology of the Chelan watershed. Two rivers were chosen from each Upper Columbia subbasin: Entiat River and Mad River (Entiat subbasin), Peshastin Creek and Chiwaukum Creek (Wenatchee subbasin), and Lost River and Early Winters River (Methow subbasin). While Chiwaukum Creek is colder and heavily forested compared to the Chelan River, it does experience summer temperatures exceeding 18°C, the temperature threshold for selecting comparison streams.

For all comparison streams, taxonomic diversity (abundance of individual taxa) and total drift biomass (g/m^3) metrics were averaged across all available years (2011-2015) and site visits (varies by river and study design panel). For the Chelan River, these metrics were averaged across both periods and all strata.

RESULTS

Summary Data

Taxonomic analysis was completed by Rhithron. Technical summary reports and raw taxonomic data are available upon request. Summary statistics were compiled showing overall abundance and taxa richness for each strata by period and collection method, strata by year, and combined for the entire river by period and year (Table 1). Taxa richness represents the number of unique taxa encountered for a sample group, so is not necessarily additive when lumping groups, for example, a taxon that occurs in two strata will be counted twice to report taxonomic richness of those 2 strata separately, but is only counted once when lumping the two strata.

Total taxonomic richness of combined benthic and drift macroinvertebrate communities was 100. Taxonomic richness was higher in the August period than May for both benthic and drift communities. Overall taxonomic richness was generally higher for the benthic community than the drift community, but abundance was orders of magnitude higher for drift than benthic. However, there was a high amount of variability in these metrics when comparing drift and benthic taxonomic diversity and abundance between strata and periods. Drift abundance was still relatively higher than benthic abundance across all strata and periods, but the combined total metrics were biased high by two outlier samples taken in the habitat channel and tailrace where a single taxon (Cladocera in Tailrace in August and Copepoda in Habitat Channel in May) dominated abundance within those samples. There were a total of 19 taxa common between drift and benthic communities, with common taxa occurring in all strata where both drift and benthic communities were sampled. This suggests that passive downstream dispersal may be an important mechanism for distribution/recruitment of the benthic community.

B-IBI scores were calculated for each strata/period (Table 2). Chelan River B-IBI were generally low, with the lowest score (14) above the dam in August (14) and the highest scores (24) in Middle Reach 1 in May and Habitat Channel in August.

Strata		Upper	Reach 1			Middle Reach 1				on Methods: D =Benthic Kick-	
Period	May Aug		Ма	May A				thic Ekman Di			
Collection Method ¹	D	BK	D	BK	D	ВК	D	BK		n Taxa occurr	
Abundance	4849	1649	583	2349	1516	343	3180	673		t and benthic s Taxa are not	samples;
Sample Taxa Richness	17	21	30	22	23	20	29	21		d in any other	strata
Common Taxa ²	4		9		8			10			
Total Taxa Richness by Period	34	1	43		35			40			
Total Taxa Richness		Ę	50			5	2				
Distinct Taxa ³			3			:	3				
Strata		Lower	Reach 1		I	Habitat	Channe	I			
Period	Ма	iy	Au	g	Ма	у	4	Aug			
Collection Method ¹	D	BK	D	BK	D	BK	D	BK			
Abundance	2746	471	4924	505	131073	1378	1741	294			
Sample Taxa Richness	25	27	24	25	11	28	27	24			
Common Taxa ²	10)	9		4			11			
Total Taxa Richness by Period	42	2	40		35			40			
Total Taxa Richness		Ę	56			5	2				
Distinct Taxa ³			5				2				
Strata		Tail	race		Ab	ove Dar	n		Combi	ned Strata	
Period	Ма	iy	Au	g	Мау	July	Aug	Ма	ay	Aug	
Collection Method ¹	D	BK	D	BK	ВК	BD	BD	D	В	D	В
Abundance	10080	382	329003	155	11	246	111	150264	4234	339431	4087
Sample Taxa Richness	15	27	3	16	7	28	14	39	49	47	50
Common Taxa ²	1		1		-	-	-	18	8	19	
Total Taxa Richness by Period	4′	1	18		-	-	-	70	C	78	
Total Taxa Richness		2	15		36					100	
Distinct Taxa ³			3			17				-	

TABLE 1. Summary table of taxa richness and abundance for drift and benthicmacroinvertebrate samples collected in Chelan River, 2016.

			May Sam	pling Period		
METRICS	R1 Upper	R1 Middle	R1 Lower	Hab Chan	Tailrace	Above Dam ¹
Taxa Richness	21	20	27	28	27	28
E Richness	1	1	2	2	0	0
P Richness	0	0	0	0	0	0
T Richness	3	4	5	3	5	1
Pollution Sensitive Richness	0	0	0	0	0	0
Clinger Richness	6	6	9	7	4	1
Semivoltine Richness	0	0	1	2	1	1
Pollution Tolerant Percent	3.00%	6.12%	9.13%	10.97%	40.58%	46.34%
Predator Percent	10.33%	9.04%	11.25%	15.54%	26.44%	7.32%
Dominant Taxa (3) Percent	74.00%	61.81%	45.44%	45.52%	56.28%	50.00%
METRIC SCORES						
Taxa Richness	3	3	3	3	3	3
E Richness	1	1	1	1	1	1
P Richness	1	1	1	1	1	1
T Richness	1	1	3	1	3	1
Pollution Sensitive Richness	1	1	1	1	1	1
Clinger Richness	1	1	1	1	1	1
Semivoltine Richness	1	1	1	1	1	1
Pollution Tolerant Percent	5	5	5	5	3	3
Predator Percent	3	1	3	3	5	1
Dominant Taxa (3) Percent	3	3	5	5	3	5
MAY SAMPLE SCORE	20	18	24	22	22	18
	20	10		mpling Period		10
METRICS	R1 Upper	R1 Middle	R1 Lower	Hab Chan	Tailrace	Above Dam
Taxa Richness	22	21	25	24	16	14
E Richness	1	1	1	1	0	0
P Richness	0	0	0	0	0	0
T Richness	3	2	3	4	1	0
Pollution Sensitive Richness	2	0	0	1	0	0
Clinger Richness	8	6	8	8	3	0
Semivoltine Richness	Ő	Ő	1	1	1	1
Pollution Tolerant Percent	2.57%	1.32%	2.97%	8.16%	10.97%	45.95%
Predator Percent	9.54%	21.55%	10.30%	22.11%	19.35%	1.80%
Dominant Taxa (3) Percent	77.25%	73.35%	60.20%	48.30%	81.29%	64.86%
METRIC SCORES			00.2070		02070	0
Taxa Richness	3	3	3	3	1	1
E Richness	1	5 1	1	1	1	1
P Richness	1	1	1	1	1	1
T Richness	1	1	1	1	1	1
Pollution Sensitive Richness	1	1	1	1	1	1
Clinger Richness	1	1	1	1	1	1
		1	1	1		1
	1	1	1	1	1	
Semivoltine Richness	1	1	1 5	1	1	3
Semivoltine Richness Pollution Tolerant Percent	1 5	1 5 5	1 5 3	1 5 5	1 5 3	1 3 1
Semivoltine Richness Pollution Tolerant Percent Predator Percent	1 5 1	5	3	5	1 5 3	1 3 1 2
Semivoltine Richness Pollution Tolerant Percent	1 5 1 1 1	-	•	-	1 5 3 <u>1</u> 16	3 1 3 14

TABLE 2. Benthic Index of Biotic Integrity (B-IBI) metrics and scores for the Chelan River,2016. Metrics calculated by Rhithron Associates, Inc.

¹Above Dam sample was taken in July. May sample was discarded from analyses.

Benthic Macroinvertebrate Community- Taxonomic Diversity

A diversity of benthic taxa was encountered in the Chelan River. There was strong differentiation of Lake Chelan (above dam) and river assemblage structures (combined periods). The benthic community structures were most similar between the Habitat Channel and Reach 1 (Upper, Middle and Lower) of the Chelan River, with some dissimilarity between these four strata and the Tailrace. The preliminary ordination analysis showed that the community structure above the dam was different to all other strata (Figure 4); however, some convergence issues persisted in our analysis due to the small sample size.

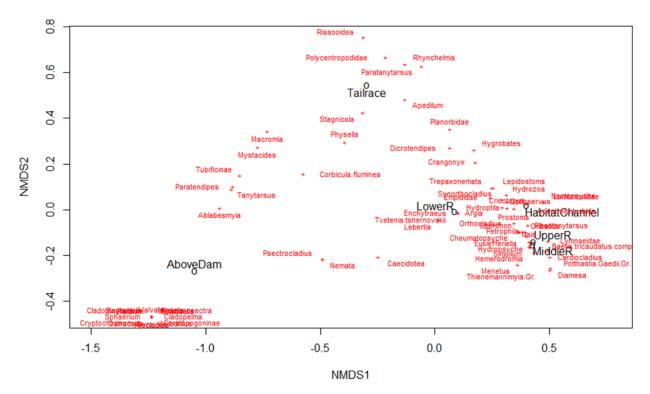


FIGURE 4. Non-metric multidimensional scaling (NMDS) ordination based on benthic taxa abundances of combined sampling periods in six sample strata (Tailrace, Lower Reach 1, Middle Reach 1, Upper Reach 1, Above Dam and Habitat Channel). Red text shows taxa.

Benthic Community Dispersal / Recruitment

The benthic community structure above and below the dam was highly dissimilar for both periods regardless of which analysis method was chosen (Bray-Curtis or Sørenson; Table 3). Using abundance data (Bray-Curtis; Clarke and Warwick 2001) the maximum similarity between community structures was only 7.32% for any strata/period below the dam and the above dam stratum; however, using taxa presence/absence (Sørenson index), similarity was somewhat higher, with a maximum similarity of 17.91% for any strata/period, except for the Tailrace which was 41.27% similar to the Above Dam stratum for combined periods. Community structures were relatively more similar between the Tailrace and all other below dam strata than the Above Dam stratum for all periods. There was a high similarity of community structures between Reach 1 and the Habitat Channel, as high as 82.4% in Reach 1 Middle in May (Sørensen; Chao *et al.* 2005). We could not test statistical significance of these results because these data are limited in only representing a single year with only a single sample for each stratum/period.

TABLE 3. Benthic similarities indices (Bray-Curtis similarities [Clarke and Warwick 2001] and Sørensen similarities indices [Chao *et al.* 2005]) in percentage between sites in the Chelan River for samples collected during May1, August and both periods combined. "HabCh", "R1Low", "R1Mid", "R1Upp" and "Tailrace" represent Habitat Channel, Lower Reach 1, Middle Reach 1, Upper Reach 1, and Tailrace, respectively.

Sites	E	Bray-Curtis	Similarities	Index (%)		Sørei	nsen Simila	rities Index	(%)	
	Ab.Dam	HabCh	R1Low	R1Mid	R1Upp	Ab.Dam	HabCh	R1Low	R1Mid	R1Upp
a. May ¹ samp	ling period									
HabCh	1.74					14.04				
R1Low	3.07	48.63				14.29	80.70			
R1Mid	1.36	37.42	63.14			8.33	73.47	70.83		
R1Upp	1.00	45.11	38.02	34.02		11.76	80.77	82.35	88.37	
Tailrace	7.32	30.92	33.76	22.34	15.53	40.00	53.57	61.82	46.81	52.00
b. August san	npling period									
HabCh	0.49					5.13				
R1Low	0.32	54.57				5.00	62.75			
R1Mid	1.88	40.67	57.58			11.43	60.87	63.83		
R1Upp	0.35	17.63	30.34	32.77		5.88	71.11	65.22	68.29	
Tailrace	1.50	18.71	11.52	8.45	3.42	13.79	50.00	48.78	33.33	40.00
c. Combined	sampling peri	ods								
HabCh	1.59					17.65				
R1Low	2.10	65.36				17.91	73.97			
R1Mid	1.80	46.70	69.89			13.33	78.79	70.77		
R1Upp	0.63	39.01	37.50	33.87		10.17	83.08	75.00	84.21	
Tailrace	6.71	28.93	24.06	19.63	10.11	41.27	60.87	61.76	59.02	56.67

¹Above Dam samples compare July/August, all other strata compare May/August. Above Dam May sample was discarded for this analysis.

Benthic Community Comparison

The Chelan River B-IBI metric scores were lower than all comparison streams. High temperature and lake-fed sites scored much higher than the Chelan River. The Palouse River scored lower than the other comparison sites, but generally scored better than the Chelan River. The Chelan River and Palouse River both showed low scores in semi-voltine and clinger metrics (Figures 5 and 6).

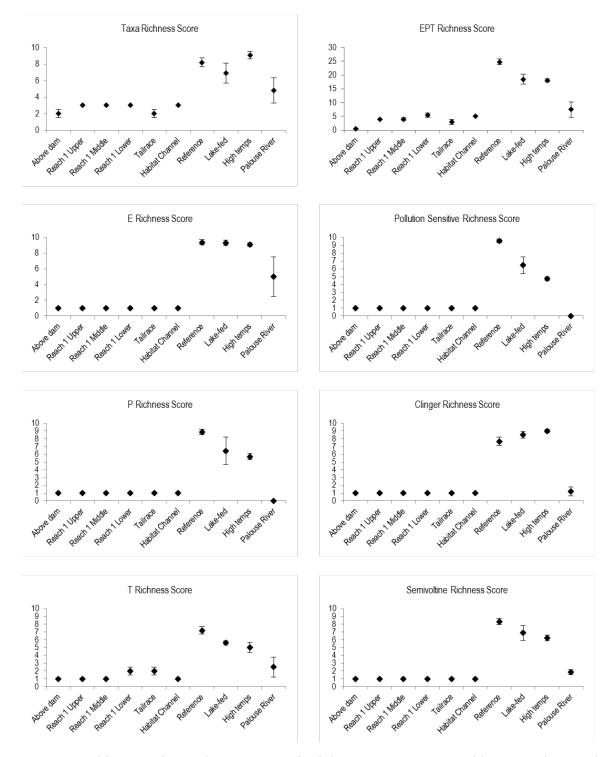


FIGURE 5. Benthic macroinvertebrate taxonomic richness scores compared between six sample reaches of the Chelan River, vs. reference and comparison stream conditions.

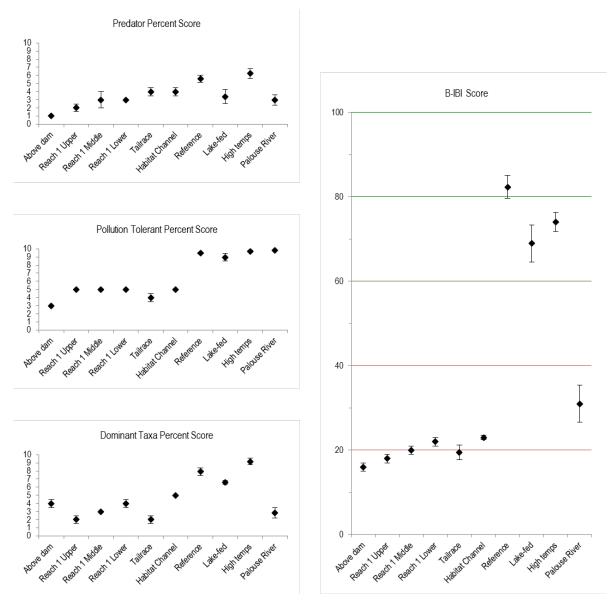


FIGURE 6. Benthic macroinvertebrate taxonomic percent and Benthic Index of Biotic Integrity (B-IBI) scores compared between six sample reaches of the Chelan River, vs. reference and comparison stream conditions. {B-IBI Scores: [0,20] = Very Poor, [20,40] = Poor, [40,60] = Fair, [60,80] = Good, [80,100] = Excellent}.

Drift Macroinvertebrate Community

Drift biomass was calculated for all samples as dry mass of drift organisms collected divided by the volume of water sampled (g/m³), which was calculated as a function of the average flow through the nets over the time period nets were deployed. In general, biomass was higher within the habitat channel and tailrace strata (Figure 7); however, these metrics were biased high by two samples with very high biomass of a single taxon (Cladocera in Tailrace in August and Copepoda in Habitat Channel in May). When looking only at Reach 1 (Figure 8),

biomass generally increased from upstream to downstream, but high variability precluded any statistical analysis of significance.

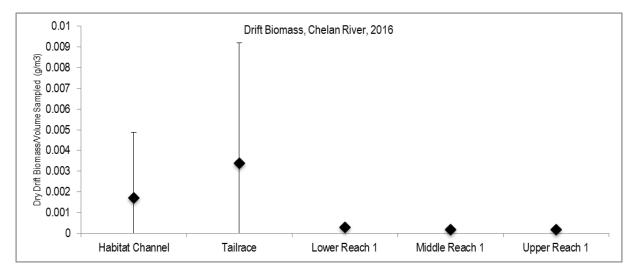


FIGURE 7. Average biomass of drift macroinvertebrate community in the Chelan River, combined periods, calculated as dry sample mass / volume sampled (g/m³).

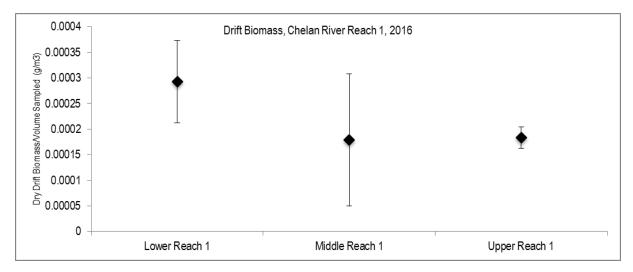


FIGURE 8. Average biomass of drift macroinvertebrate community in Reach 1 of the Chelan River, combined periods, calculated as dry sample mass / volume sampled (g/m³).

Drift macroinvertebrates were categorized by resource category (group) as aquatic, terrestrial, or aquatic-terrestrial, and presented as proportionate abundance within each strata/period (Figure 9). Aquatic taxa exclusively inhabit aquatic habitats. Terrestrial taxa exclusively inhabit terrestrial environmental habitats and are essentially incidental to drift samples, but offer an important nutrient source for predators and overall water quality. Aquatic-terrestrial taxa are the terrestrial adult life stage of taxa that occupy both aquatic and terrestrial habitats during different life stages. Taxa of this category may be included in both aquatic and aquatic-terrestrial taxonomic groups within the same sample because they occur in varying life stages. In general, abundances of aquatic taxa were higher in the tailrace and habitat channel strata for both periods than in the Reach 1 strata, although again, these groups may be biased high by outlier samples as discussed previously. Within Reach 1, aquatic taxa had higher abundance in August than May, and combined aquatic and aquatic-terrestrial groups had higher abundance in all strata and periods than terrestrial taxa.

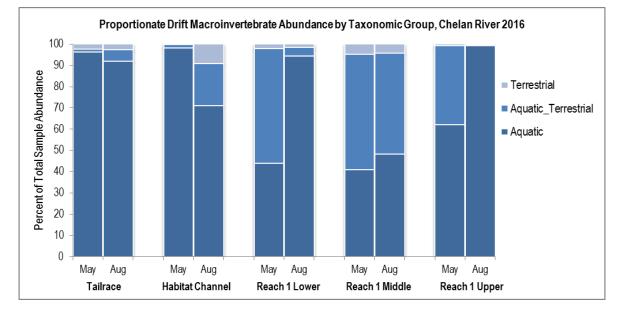


FIGURE 9. Proportionate abundance of drift macroinvertebrate taxa by group (terrestrial, aquatic, aquatic-terrestrial) within the Chelan River by strata and period, 2016.

The drift macroinvertebrate communities had greater similarity between Reach 1 and the Habitat Channel, than the Tailrace and other strata (Figure 10). Taxa shown closer together indicate greater similarity in composition between strata than those further apart. For example, the taxa Trichoptera and Physidae were present only in Upper Reach 1, whereas Leptoceridae and Hydroptidae were found more in the Habitat Channel than other strata. The stress value (goodness of fit) of the analysis was almost zero, similar to the benthic NMDS analysis. This analysis would be improved with additional data.

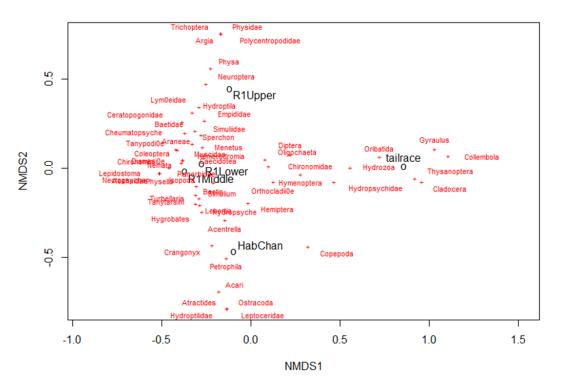


FIGURE 10. Non-metric multidimensional scaling (NMDS) ordination based on species density in 5 sample sites (Tailrace, Lower Reach 1, Middle Reach 1, Upper Reach 1 and Habitat Channel). Taxa are in red text.

Drift Community Dispersal / Recruitment

We found a high level of variability among drift macroinvertebrate community structure between strata, taxonomic groups and periods (Table 4). Generally, there was a greater similarity in both abundance (Bray-Curtis; Clarke and Warwick 2001) and taxa presence/absence (Sørenson; Chao *et al.* 2005) between the three sections of Reach 1 and the habitat channel, particularly for aquatic taxa. The taxonomic composition (without abundance) was as high as 70% for aquatic grouped taxa in May. We did not collect drift samples above the dam, and so cannot compare drift communities above and below the dam.

Using abundance data (Bray-Curtis; Clarke and Warwick 2001) for combined periods and groups, the highest similarity between any strata was between the middle and lower sections of Reach 1 (63.61%). In terms of taxa presence/absence (Sørenson) for combined periods and groups, the highest similarity was between upper and lower Reach 1 (75.68%). However, similarity for this metric exceeded 50% between all strata combinations except lower Reach 1 and the Tailrace (40%). It may not be appropriate to combine strata and groups for drift macroinvertebrate similarity indices considering the high level of variability in these metrics, but increasing the time series of this dataset could reduce noise and allow greater statistical power in analyzing trends. We could not test statistical significance of these results because these data are limited in only representing a single year with only a single sample for each stratum/period. TABLE 4. Drift similarities indices (Bray-Curtis [Clarke and Warwick 2001] and Sørensen [Chao *et al.* 2005] similarities indices) in percentage between sites in the Chelan River for samples collected during May, August and both periods combined. "HabCh", "R1Low", "R1Mid", "R1Upp" and "Tailrace" represent Habitat Channel, Lower Reach 1, Middle Reach 1, Upper Reach 1, and Tailrace, respectively.

a. May 2016		Bray-	Curtis Simil	Søre	Sørensen Similarities Index (%)				
Group ¹	Strata	HabCh	Tailrace	R1Low	R1Mid	HabCh	Tailrace	R1Low	R1Mid
	Tailrace	13.76				70			
Aquatia	R1Low	1.58	8.72			41.38	41.38		
Aquatic	R1Mid	0.84	5.79	65.46		51.85	51.85	72.22	
	R1Upp	4.15	38.13	30.5	26.12	66.67	58.33	54.55	58.06
	Tailrace	13.43				100			
Aquatia torractrial	R1Low	86.22	16.67			40	40		
Aquatic-terrestrial	R1Mid	60.14	28.24	71.21		66.67	66.67	66.67	
	R1Upp	94.44	13.98	90.34	62.74	33.33	33.33	88.89	57.14
	Tailrace	4.87				33.33			
	R1Low	6.06	23.89			33.33	60		
Terrestrial	R1Mid	14.7	21.09	60.32		28.57	72.73	72.73	
	R1Upp	7.25	21.58	48.35	48.65	50	50	50	44.44
	Tailrace	13.72				61.54			
.	R1Low	3.72	10.07			36.84	42.86		
Combined	R1Mid	2.1	8.05	68.42		45.71	56.41	70.59	
	R1Upp	6.6	34.7	56.56	41.57	58.06	51.43	59.57	54.55
b. August 2016	кторр	0.0	04.7	00.00	41.07	00.00	01.40	00.07	04.00
5.7 August 2010	Tailrace	0				8			
	R1Low	54.36	0			65.12	0		
Aquatic	R1Mid	60.67	0	63.81		59.57	Ö	77.27	
	R1Upp	26.02	0 ²	12.53	15.85	68.09	7.69	72.73	66.67
	Tailrace	23.15		12.00	10.00	33.33	1.00	12.10	
	R1Low	17.28	68.12			88.89	40		
Aquatic-terrestrial	R1Mid	80	26.73	22.26		80	33.33	88.89	
	R1Upp	49.46	24.16	26.46	61.65	80	33.33	88.89	100
	Tailrace	0	24.10	20.40	01.00	0	00.00	00.00	100
	R1Low	15.29	0			66.67	0		
Terrestrial	R1Mid	65.17	0	14.06		88.89	0	60	
	R1Upp	40.58	0	9.62	68.57	66.67	0	60	80
	Tailrace	0.11	0	9.02	00.07	12.12	0	00	
	R1Low	45.7	0.3			72.41	6.45		
Combined	R1Low R1Mid	45.7 61.78	0.3	55.73		67.74	6.45 5.71	76.67	
		32.53	0.16	55.73 15.43	22.54	67.74	5.71 11.43	73.33	71.88
a Combined Derieda	R1Upp	32.00	0.07	10.40	22.04	07.74	11.43	13.33	/ 1.00
c. Combined Periods	Toilroos	01 00				E1 06			
	Tailrace	24.32	07			51.06	40		
Combined	R1Low	6.98	0.7	00.04		65.67	40	00.44	
	R1Mid	5.05	0.68	63.61	45.0.1	66.67	46.15	69.44	
	R1Upp	7.21	1.84	45.7	45.24	67.61	48.15	75.68	68.42

¹Some taxa such as Chironomidae may be categorized in both aquatic-terrestrial and aquatic groups, as different life stages occupy different habitats. For example, Chironomidae larvae are categorized as aquatic, and Chironomidae adults are categorized as aquatic-terrestrial. Therefore, total taxa richness value may vary slightly between analyses depending on how groups are split or lumped, and total taxa richness of combined groups may be less than the sum of taxa richness within groups.

²The tailrace has a large abundance of Cladocera (32,7001), resulting in community dissimilarity. Bray-Curtis similarities take into account species type and abundance, whereas Sørensen only accounts for species.

Drift Community Comparison

We compared community composition using overall abundance of the 20 dominant drift taxa (combined groups and periods) encountered in the Chelan River with six comparison streams in the Wenatchee, Entiat and Methow subbasins, chosen from the CHaMP database for similar discharge and channel width profiles. The Chelan River was largely dominated by Cladocera and Copepoda (Figure 11), which were non-existent or extremely rare in all comparison streams. Further investigation shows that these taxa were encountered in extremely high numbers in only two samples in the Chelan River, with Copepoda found only in the May Habitat Channel sample, and Cladocera found only in the August Tailrace sample. If these outliers are removed from the biodiversity comparison (Figure 12), we see a much greater similarity between taxa found in the Chelan River and comparison streams. The three most dominant taxa in the Chelan River, Chironomidae, Baetis and Orthocladinae, were also found in high abundances in all comparison streams except Chiwaukum, which is higher elevation and more shaded than any of the other comparison streams.

We compared total collected biomass of drift macroinvertebrates with the same CHaMP comparison streams (Figure 13), and found that the Chelan River showed generally higher biomass than the comparison streams, with poor confidence in the estimate. However, considering the same possible bias from high Cladocera and Copepoda presence in two samples of the confluence reaches, we also compared biomass between only Reach 1 strata and the comparison streams (Figure 14). This comparison showed much greater similarity in biomass between Chelan River and the other streams, and a much more precise estimate of average biomass.

We also investigated percentage similarity in taxonomic composition (taxa presence/absence) between the Chelan River and the six comparison streams using Sørenson Similarities Indices (Table 5). We could not compare abundance/dominance of taxa using Bray-Curtis (Clarke and Warwick 2001) because of a high variability in sample size between all streams. We saw a range of similarity in composition between the Chelan River and comparison streams from 27% (Entiat River) to 49% (Lost River, Methow). This was similar to the range in similarities seen amongst the individual comparison streams (24 - 70%). We could not test statistical significance of biomass or taxa richness results because the Chelan River data are limited in only representing a single year with only a single sample for each stratum/period.

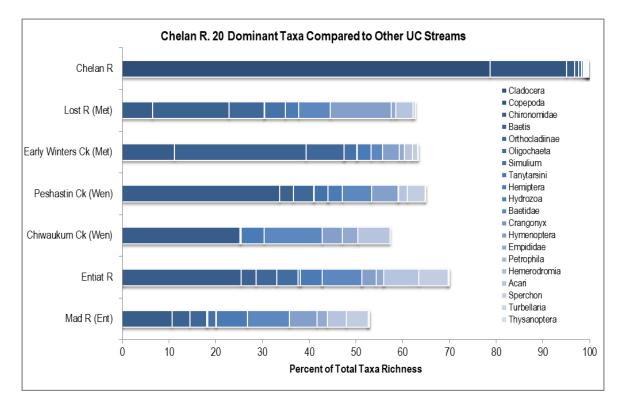


FIGURE 11. Overall abundance of the 20 dominant drift macroinvertebrate taxa encountered in the Chelan River (combined taxonomic groups, periods and strata) in 2016, compared with 6 similar streams in the Upper Columbia region. Comparison streams were chosen from the CHaMP database for similar discharge and channel width profiles, and combine data from many sites and years (2011 - 2015) within each stream.

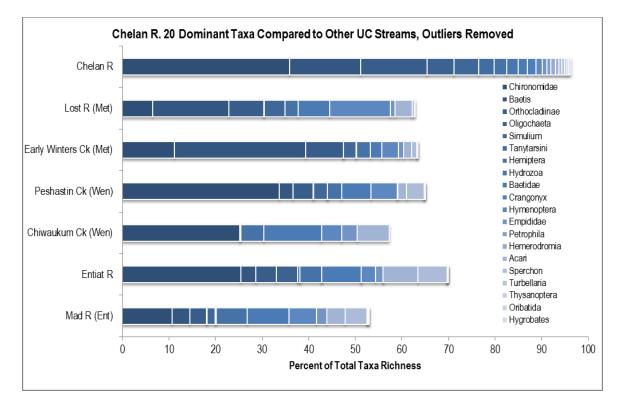


FIGURE 12. Overall abundance of the 20 dominant drift macroinvertebrate taxa, not including Cladocera or Copepoda, encountered in the Chelan River (combined taxonomic groups, periods and strata) in 2016, compared with 6 similar streams in the Upper Columbia region. Comparison streams were chosen from the CHaMP database for similar discharge and channel width profiles, and combine data from many sites and years (2011 - 2015) within each stream.

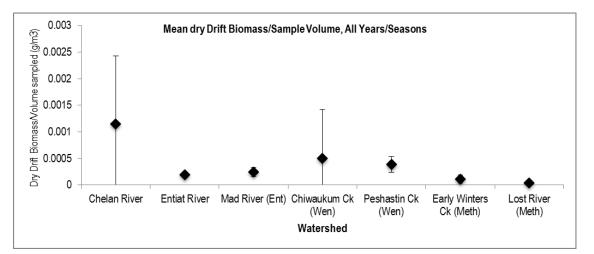
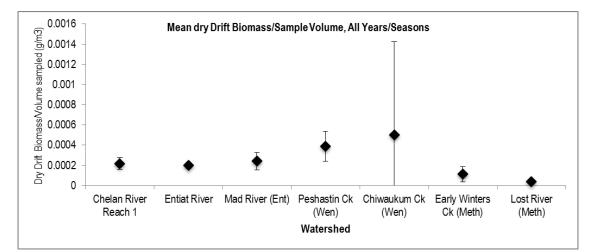


FIGURE 13. Total drift fauna biomass per sampled water volume (g/m³) collected in the Chelan River (combined taxonomic groups, periods and strata) in 2016, compared with 6 similar streams in the Upper Columbia region. Comparison streams were chosen from the CHaMP database for similar discharge and channel width profiles, and combine data from many sites and years (2011 - 2015) within each stream.



- FIGURE 14. Total drift fauna biomass per sampled water volume (g/m³) collected in Reach 1 of the Chelan River (combined taxonomic groups and periods) in 2016, compared with 6 similar streams in the Upper Columbia region. Comparison streams were chosen from the CHaMP database for similar discharge and channel width profiles, and combine data from many sites and years (2011 2015) within each stream.
- TABLE 5. Sørensen similarities indices (Chao *et al.* 2005) in percentages between the Chelan River and six comparison streams in the Wenatchee (Chiwaukum, Peshastin), Entiat (Entiat, Mad) and Methow (Lost, Early Winters) River Subbasins. Chelan River data pools all strata/periods from 2016 drift sampling. Comparison streams pool all sites (varies) and years (2011 - 2015) available from the CHaMP database.

Reference_Site	Chelan R	Mad R	Entiat R	Chiwaukum Ck	Early Winters Ck	Lost R
Mad R	27.397					
Entiat R	36.879	70.614				
Chiwaukum Ck	30.435	24.060	25.781			
Early Winters Ck	41.830	52.599	52.997	47.244		
Lost R	49.573	38.488	39.146	37.363	55.263	•
Peshastin Ck	37.037	61.708	59.490	36.810	61.607	50.000

Organic Drift Detritus

We analyzed the biomass of organic drift detritus in each strata of the Chelan River, as Ash Free Dry Mass/Weight (AFDW) by volume of water sampled (g/m³; Figure 15). There was overall a greater amount of drift detritus collected in May than there was in August, except within the upper and middle sections of Reach 1. There was a similar biomass collected in all strata except for the tailrace, where we also saw the greatest seasonal variability.

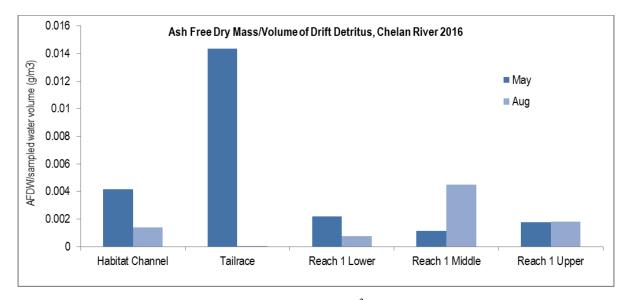


FIGURE 15. Total mass per sampled water volume (g/m³) of organic drift detritus in the Chelan River, 2016.

DISCUSSION

The field data collection component of this project was completed without any significant protocol modifications, except for a methodology change from modified kick net sampling to dredge sampling in the Above Dam stratum following poor data quality obtained in the May sample. This change yielded much greater quality data than the initially employed approach, in both taxonomic diversity and total abundance of organisms collected, so the revised protocol was permanently changed in the final Quality Assurance Project Plan (QAPP) and recommended for any future sampling efforts under this scope.

The May sampling event was logistically constrained by high flows and a necessary ramp down in dam spill leading up to sampling, which was successfully orchestrated by the PUD and allowed sampling to proceed with no issues. However, macroinvertebrate community composition could have been altered by the high flows prior to sampling, and in fact any bias could have extended into August sampling for long-lived species since flows were high shortly after the May sample period. It is possible that some benthic taxa could have been flushed out of the system by high flows, and that some new drift taxa could have potentially been introduced from the lake, but there is no way to test these hypotheses without extending the time series of sampling, and it is difficult to define baseline conditions without first accounting for natural or introduced variability in the system. We therefore recommend continuing this investigation until inter-annual variability can be adequately described.

Benthic Macroinvertebrate Community- Taxonomic Diversity

The biological diversity of the benthic macroinvertebrate community, as measured by B-IBI, was generally very low compared to reference conditions, and much lower than comparable streams in this ecoregion identified by water temperature, discharge profiles and water source.

However, in absence of pre-dam baseline conditions, it is impossible to say how much benthic community diversity would have existed with natural flows from the hyper-oligotrophic headwater conditions. Furthermore, pre-SA the river was typically dry most of the year, so any pre-dam macroinvertebrates would likely have been entirely extirpated, leaving a "clean slate" by the time the SA was implemented approximately 7 years ago. It is likely that elevated summer temperature is one factor affecting taxa richness, but that is probably not the only factor, and warm water temperatures at the outlet of Lake Chelan are a natural consequence of the shallow basin upstream of the lake outlet. Further study is needed in order to describe temporal variability and trends in the Chelan River benthic community before specific limiting factors can be determined that might be possible to mitigate. It is possible that community diversity will improve as riparian cover is further developed. The SA does not explicitly recommend any other enhancements that would be likely to influence benthic community diversity, and given the high temperature water outflowing Lake Chelan, it would be very difficult to affect a change in summer water temperatures in the river through riparian enhancement alone.

Benthic Community Dispersal / Recruitment

Similarities indices suggest that the Above Dam stratum may contribute more benthic organisms to the Tailrace stratum than to the Reach 1 or Habitat Channel strata. However, this could also be a result of the available habitat being more similar between the Tailrace and Above Dam strata (sandy substrate, low velocity). The Habitat Channel also receives 60-80% of its water pumped directly from the tailrace in the spring and fall, but the high similarity of community structures between Reach 1 and the Habitat Channel, as high as 82.4% in Reach 1 Middle in May (Sørensen; Chao *et al.* 2005) suggest that downstream dispersal from the river is a more important recruitment mechanism than from the lake to the Habitat Channel despite the seasonal direct water source.

The relative contribution of Lake Chelan (Above Dam) to the Chelan River benthic community seems to be low. This stratum also exhibited the lowest diversity of any strata sampled, a reasonable result given that any source colonies in tributaries to the lake would be unlikely to survive the distance or depth of the lake in transport to the outlet. It would therefore be unlikely to expect much colonization to the Chelan River benthic community from downstream dispersal, either via spillway to Reach 1 or powerhouse outflow to the tailrace or habitat channel. The Rocky Reach reservoir has also shown poor benthic diversity of EPT¹ taxa in previous studies (CPUD 2000), and offers a limited source for upstream migration of potential colonizers to the tailrace or habitat channel. Upstream migration would be inhibited between the Habitat Channel and Reach 1 by the Chelan River Gorge, and would be impossible through the spillway or powerhouse outflows to Lake Chelan. This means that any colonization within the Chelan River is likely dominated by aerial (e.g., wind, aerial plankton; see Bilton *et al.* 2001) or passive (e.g., waterfowl) dispersal mechanisms from distant populations, and any level of restoration within the river could be ineffective at improving benthic communities (Brederveld *et al.* 2011). Furthermore, there was a lack of Plecoptera (stoneflies) species in all strata and both

¹ Ephemeroptera (mayflies), Plecoptera (stoneflies), and Trichoptera (caddisflies), three taxa of aquatic macroinvertebrates that are intolerant to pollution and poor water quality.

seasons. Stoneflies typically occupy areas of substrate similar to the Chelan River, and are generally tolerant of short-term exposure to high water temperatures. We would therefore expect to see this taxa, at least in some numbers, within our samples if a recruitment source were available. Stoneflies are weak fliers, and therefore can only recruit from nearby streams, further supporting the hypothesis that dispersal/recruitment is the key limiting factor to colonization in the Chelan River (Briers *et al.* 2002). It is possible that translocation of EPT taxa from another stream could help establish a population within the Chelan River, but the literature is weak on this method and further study would be needed.

Benthic Community Comparison

Low B-IBI metric scores in the Chelan River, relative to all comparison streams, suggests that high summer temperatures and lake source flows cannot alone explain the low benthic community diversity of the Chelan River. The Palouse River was the closest match to the Chelan River in both discharge and temperature, and experiences similarly wide fluctuations in high to low discharge, although the seasonality of these hydrologic events differs. Of particular interest are the Chelan River and Palouse River's similarly low scores in semi-voltine and clinger metrics. Semi-voltine are long-lived species that take more than a year to complete their life cycle. They are especially sensitive to streams that run dry or have large flooding events, which is characteristic of both the Chelan and Palouse Rivers. Clingers prefer cobble and boulder habitats and are negatively impacted by high embeddedness. The Palouse River has fine sedimentation issues that are probably also affecting clinger populations, but this does not appear to be a limiting factor in the Chelan River. It therefore appears that high discharge variability is the most significant common factor influencing reduced benthic diversity between these systems, but is likely not as significant a limiting factor as biotic recruitment sources. There are many examples of other rivers that experience significant seasonal flood events and still support healthy benthic macroinvertebrate communities. Like temperature, hydrology in the Chelan River would be very difficult to control through any reasonable management actions or habitat enhancements, given its incised channel morphology, lack of sediment source, and complexity in lake level management regimes.

Drift Community Dispersal / Recruitment

Similar to benthic taxa, we showed that a greater similarity between Reach 1 and the Habitat Channel communities suggests downstream dispersal is an important recruitment mechanism, but could not directly compare drift communities above and below the dam to assess whether Lake Chelan provides adequate recruitment stock to colonize the river. Sampling in 2016 was not targeted at drift macroinvertebrates above the dam, because the initial objective was to compare taxonomy of the benthic communities above and below the dam to explore possible recruitment sources for benthic macroinvertebrates in the river. Therefore, the best comparison possible for drift macroinvertebrates above and below the dam was to use the tailrace as a proxy for the above dam environment, as it is a direct outflow, versus the other sample strata that bypass through a variety of habitats and possible recruitment sources. However, because of available habitat within protocol standards, the tailrace sample in May was located below the confluence of the habitat channel, and although flow appeared to be dominated by water from the tailrace, could have been mixed with other sources. We believe that the low

similarity in taxa abundances and relatively lower similarity in taxa compositions between the tailrace and other upstream strata suggest that, similar to the benthic community, Lake Chelan likely contributes little to the drift macroinvertebrate community in the Chelan River. However, this could warrant further investigation if organisms found closer to the lake surface are entering Reach 1 of the river via the spillway in greater quantities than are entering the tailrace through the powerhouse outflow, which could also explain some of the dissimilarity in community composition between the tailrace and other strata. This could also help further explain recruitment pathways for benthic taxa entering the river via spillway from Lake Chelan if they are waterborne and not accessible to collection by benthic kicknet in Reach 1. We recommend development and testing of a method such as plankton trawl nets to allow drift macroinvertebrate sampling for comparison above the dam. The developed method must allow measurement of sample volume, and sample a similar volume to nets set in wadeable strata so that biomass and diversity can be directly compared.

Drift Community Comparison

Biomass and taxonomic composition of the drift community were at a similar level to the six Upper Columbia CHaMP comparison streams. Overall taxonomic diversity was somewhat lower, but without other lake-fed streams available for comparison it is difficult to draw any conclusions from this. Although further bioenergetics modeling would be needed, it is likely that drift macroinvertebrates provide an important food source for resident and rearing fish in Reach 1 and the Habitat Channel.

Outlier samples of Cladocera (May Habitat Channel) and Copepoda (August Tailrace) were found in only two samples in the Chelan River and virtually nowhere else in comparison streams. These taxa are also uncommon in the Columbia River, but have been found in previous plankton surveys in Lake Chelan (S. Hays pers. comm.). It is therefore likely these species were entrained at the Chelan Dam penstock intake. Habitat Channel flows in May were supplemented by pumped tailrace water, so the lack of copepods in the May tailrace sample is puzzling; however, it is not uncommon to observe patchy distribution of zooplankton, particularly in deep, slow moving water. The lack of Cladocera in the Habitat Channel in August is not surprising since there was no pumped tailrace water at that time of year. The presence of Copepoda and Cladocera taxa may not accurately represent the biodiversity of the Chelan River, but they could provide an important food source for fish rearing in the tailrace and habitat channel. It is somewhat surprising that neither of these lake-sourced taxa were found in the Reach 1 samples, but perhaps because the low-level outlet draws water only from the bottom 18 inches at the Chelan Dam, there is not as much exchange of pelagic organisms entrained by the low-level outlet as the penstock.

Organic Drift Detritus

Overall, we observed a greater amount of drift detritus in May than in August, which is contrary to expectations since increases of instream algae typically correlate with increased summer temperatures. We also saw a similar biomass in all strata except for the tailrace, where we saw the greatest seasonal variability and highest amount in the May sample. The cause of this is uncertain, but could be a result of settled detritus in Lake Chelan being upwelled and

transported through the penstock during activation of the spill overflow channel during the lead up to the May sampling window. Although turbidity was not explicitly measured, visibility seemed slightly poorer in May than August, especially within the tailrace, which could also be a result of this activation. Anecdotally, we observed a greater presence of Didymo in the upper part of Reach 1 than elsewhere, with an increase in August. This could account for some of the increase in detritus in Reach 1 in August, but it is difficult to validate as detritus was only weighed and not identified. It is unlikely that samples were influenced by leaf litter or woody debris as riparian cover is poor throughout middle and upper Reach 1, and water flowing from the lake is generally clean.

Recommendations

- 1) Repeat this study for an additional 2 years, to account for natural interannual variability in community structures, as well as any potential bias introduced by unusual flow regimes in 2016.
- 2) Introduce drift macroinvertebrate trawl sampling methods above the dam, to further explore this area as a recruitment source for downstream communities.
- 3) Increase the total sample size in future collection efforts (i.e., collect more samples within each strata or increased periodicity of sampling) to allow more advanced statistical comparison of samples.

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APPENDIX A: DRIFT TRANSECT MASTER SAMPLE LIST

SiteID	Strata	Lat	Long	Use Order	Evaluation
CRPUD16_76	Habitat Channel	47.80870702	-119.9843489	1	Accept
CRPUD16_70	Habitat Channel	47.80658178	-119.9857635	2	
CRPUD16_69	Habitat Channel	47.8061344	-119.9856961	3	Reject- too close to targeted tailrace sample
CRPUD16_72	Habitat Channel	47.80736273	-119.9854115	4	
CRPUD16_74	Habitat Channel	47.80811289	-119.9850012	5	
CRPUD16_78	Habitat Channel	47.80957276	-119.9845781	6	
CRPUD16_75	Habitat Channel	47.8082989	-119.9844046	7	
CRPUD16_79	Habitat Channel	47.81001609	-119.9846393	8	Reject- overlaps LWD structure
CRPUD16_71	Habitat Channel	47.80694381	-119.985442	9	
CRPUD16_73	Habitat Channel	47.80780206	-119.985429	10	
CRPUD16_77	Habitat Channel	47.80912313	-119.9845803	11	
CRPUD16_68	Habitat Channel	47.80569071	-119.9855889	12	Reject- too close to targeted tailrace sample
CRPUD16_18	Reach 1 Lower	47.82394305	-119.9962547	1	Accept
CRPUD16_21	Reach 1 Lower	47.82446537	-119.998092	2	
CRPUD16_20	Reach 1 Lower	47.82429593	-119.9974814	3	
CRPUD16_09	Reach 1 Lower	47.82254983	-119.9906717	4	
CRPUD16_15	Reach 1 Lower	47.82346597	-119.9943936	5	
CRPUD16_10	Reach 1 Lower	47.82263946	-119.9913255	6	
CRPUD16_01	Reach 1 Lower	47.81959611	-119.9887145	7	
CRPUD16_12	Reach 1 Lower	47.82287537	-119.9925997	8	
CRPUD16_04	Reach 1 Lower	47.82089452	-119.9886463	9	
CRPUD16_22	Reach 1 Lower	47.82475478	-119.9985928	10	
CRPUD16_08	Reach 1 Lower	47.82237125	-119.9900644	11 12	
CRPUD16_14	Reach 1 Lower	47.82330034	-119.9937734		
CRPUD16_16	Reach 1 Lower	47.82365456	-119.9949979	13	
CRPUD16_05 CRPUD16_07	Reach 1 Lower	47.82131948 47.82208843	-119.9888588	14 15	
CRPUD16_07 CRPUD16_19	Reach 1 Lower Reach 1 Lower	47.82208843	-119.9895468 -119.9968667	15	
CRPUD16_03	Reach 1 Lower	47.8204676	-119.9908007	10	
CRPUD16_06	Reach 1 Lower	47.82170925	-119.9891906	17	
CRPUD16_00	Reach 1 Lower	47.82378921	-119.9956346	18	
CRPUD16_13	Reach 1 Lower	47.82309888	-119.9931782	20	
CRPUD16_11	Reach 1 Lower	47.82270277	-119.9919863	20	
CRPUD16_02	Reach 1 Lower	47.82002174	-119.9885045	22	
CRPUD16_28	Reach 1 Middle	47.8273328	-119.9984114	1	Accept
CRPUD16 43	Reach 1 Middle	47.83216519	-120.0001326	2	
CRPUD16_29	Reach 1 Middle	47.82775152	-119.9981699	3	
CRPUD16_31	Reach 1 Middle	47.82862035	-119.9978341	4	
CRPUD16_23	Reach 1 Middle	47.82515805	-119.9988705	5	
CRPUD16 26	Reach 1 Middle	47.82649323	-119.9988818	6	
CRPUD16_35	Reach 1 Middle	47.83037736	-119.9972999	7	
CRPUD16_36	Reach 1 Middle	47.83082639	-119.9972741	8	
CRPUD16_25	Reach 1 Middle	47.82604891	-119.998971	9	
CRPUD16_42	Reach 1 Middle	47.83235707	-119.9995334	10	
CRPUD16_44	Reach 1 Middle	47.83193357	-120.0007049	11	
CRPUD16_32	Reach 1 Middle	47.82905871	-119.9976857	12	
CRPUD16_30	Reach 1 Middle	47.82818019	-119.99797	13	
CRPUD16_41	Reach 1 Middle	47.83244431	-119.9988826	14	
CRPUD16_27	Reach 1 Middle	47.82692022	-119.9986763	15	
CRPUD16_40	Reach 1 Middle	47.83235911	-119.9982334	16	
CRPUD16_34	Reach 1 Middle	47.82993417	-119.9973909	17	
CRPUD16_38	Reach 1 Middle	47.83171723	-119.9973497	18	
CRPUD16_33	Reach 1 Middle	47.82949871	-119.9975491	19	
CRPUD16_37	Reach 1 Middle	47.83127585	-119.9972586	20	
CRPUD16_24	Reach 1 Middle	47.82559939	-119.99898	21	
CRPUD16_39	Reach 1 Middle	47.83209356	-119.9977055	22	
CRPUD16_51	Reach 1 Upper	47.83313926	-120.0031799	1	Reject- Field Eval, visible fire retardant prevalent in site
CRPUD16_59	Reach 1 Upper	47.83632567	-120.0052844	2	Accept
CRPUD16_60	Reach 1 Upper	47.83666325	-120.0056976	3	
CRPUD16_49	Reach 1 Upper	47.83224225	-120.0031569	4	
CRPUD16_47	Reach 1 Upper	47.83148244	-120.0025632	5	

SiteID	Strata	Lat	Long	Use Order	Evaluation
CRPUD16_65	Reach 1 Upper	47.83571112	-120.0080095	6	
CRPUD16_45	Reach 1 Upper	47.83171421	-120.001287	7	
CRPUD16_58	Reach 1 Upper	47.83596184	-120.0049004	8	
CRPUD16_62	Reach 1 Upper	47.83678706	-120.0069083	9	
CRPUD16_66	Reach 1 Upper	47.83529514	-120.0082591	10	
CRPUD16_54	Reach 1 Upper	47.83446164	-120.0034543	11	
CRPUD16_67	Reach 1 Upper	47.83486163	-120.008461	12	Reject- too close to top of survey extent
CRPUD16_64	Reach 1 Upper	47.83612871	-120.007763	13	
CRPUD16_46	Reach 1 Upper	47.83155291	-120.0019094	14	
CRPUD16_57	Reach 1 Upper	47.83557471	-120.0045663	15	
CRPUD16_50	Reach 1 Upper	47.8326898	-120.0031948	16	
CRPUD16_48	Reach 1 Upper	47.83181062	-120.0029809	17	
CRPUD16_55	Reach 1 Upper	47.83483914	-120.0038108	18	
CRPUD16_63	Reach 1 Upper	47.83650824	-120.0074289	19	
CRPUD16_53	Reach 1 Upper	47.83403419	-120.0032564	20	
CRPUD16_56	Reach 1 Upper	47.83519426	-120.0042203	21	
CRPUD16_61	Reach 1 Upper	47.83687645	-120.0062787	22	
CRPUD16_52	Reach 1 Upper	47.83358865	-120.0031889	23	

APPENDIX B: BENTHIC REFERENCE AND COMPARISON STREAM DATA

Site ID	Stream Name	Comparison Group	Taxa Richness	Taxa Richness Score	E Richness	E Richness Score	P Richness	P Richness Score	T Richness	T Richness Score	Pollution Sensitive Richness	Pollution Sensitive Richness Score
BIO06600- ROBI77	Robinson Creek	Reference Site	41	4.8	8	10	8	10	7	7.5	9	10
BIO06600- TRCO09	Tributary to Coleman Creek	Reference Site	30	1	4	4.3	7	8.6	2	1.2	4	5.7
SEN06600- TWEN05	Twentyfive Mile Creek	Reference Site	60	10	12	10	10	10	10	10	11	10
SEN06600- TEAN04	Teanaway River Middle Fork	Reference Site	61	10	10	10	4	4.3	7	7.5	7	10
BIO06600- WILS09	Wilson Creek	Reference Site	57	10	10	10	10	10	6	6.2	11	10
BIO06600- FIRS09	First Creek	Reference Site	57	10	9	10	10	10	6	6.2	7	10
BIO06600- SHAD09	Shadow Creek	Reference Site	55	9.7	9	10	8	10	6	6.2	10	10
BIO06600- NTAN09	Taneum Creek, North Fork	Reference Site	57	10	13	10	6	7.1	9	10	12	10
BIO06600- STAN09	Taneum Creek, South Fork	Reference Site	51	8.3	12	10	8	10	12	10	14	10

Site ID	Stream Name	Comparison Group	Taxa Richness	Taxa Richness Score	E Richness	E Richness Score	P Richness	P Richness Score	T Richness	T Richness Score	Pollution Sensitive Richness	Pollution Sensitive Richness Score
WAM06600- 008478	Yakima River	Lake-fed	54	9.3	7	8.6	3	2.9	5	5	3	4.3
SEN06600- CLEE12	Cle Elum River	Lake-fed	40	4.5	8	10	8	10	6	6.2	6	8.6
WAM06600- 000586	Palouse River	Best Match	32	1.7	1	0	0	0	1	0	0	0
BIO06600- SFCO10	Cowiche River, South Fork	High Temperature	48	7.2	7	8.6	5	5.7	4	3.8	3	4.3
SEN06600- UMTA18	Umtanum Creek	High Temperature	56	10	7	8.6	4	4.3	7	7.5	3	4.3
WAM06600- 009134	Teanaway River West Fork	High Temperature	58	10	10	10	6	7.1	4	3.8	4	5.7

Site ID	Clinger Richness	Clinger Richness Score	Semivoltine Richness	Semivoltine Richness Score	Pollution Tolerant Percent	Pollution Tolerant Percent Score	Predator Percent	Predator Percent Score	Dominant Taxa Percent	Dominant Taxa Percent Score	EPT Richness	IBI
BIO06600- ROBI77	17	5.9	5	3.8	0.6	9.9	7.2	3.1	43.4	6.9	23	71.8
BIO06600- TRCO09	6	0	9	8.8	4.4	9	7.4	1.7	62.8	1.7	13	43.5
SEN06600- TWEN05	26	10	9	8.8	4.2	9	9.6	4.3	38.2	8.3	32	90.4
SEN06600- TEAN04	26	10	6	5	3.6	9.2	12.6	5.8	28.8	10	21	81.7
BIO06600- WILS09	20	7.6	11	10	1	9.8	15.4	7.2	31	10	26	90.9
BIO06600- FIRS09	21	8.2	11	10	2.2	9.5	14	6.5	27	10	25	90.5
BIO06600- SHAD09	19	7.1	11	10	0.6	9.9	15.2	7.1	46.4	6.1	23	86
BIO06600- NTAN09	28	10	9	8.8	0	10	10.2	4.6	39	8.1	28	88.6
BIO06600- STAN09	29	10	12	10	4.9	8.9	43.7	10	29.4	10	32	97.1
WAM06600- 008478	23	9.4	6	5	8.6	8	4.4	1.7	46.6	6.1	15	60.2
SEN06600- CLEE12	20	7.6	9	8.8	0.6	9.9	11.2	5.1	42.8	7.1	22	77.7
WAM06600- 000586	7	0	4	2.5	1.4	9.7	9.4	4.2	53.9	4.1	2	22.2
BIO06600- SFCO10	21	8.2	6	5	0.2	10	10.6	4.8	41.2	7.5	16	65.1
SEN06600- UMTA18	22	8.8	8	7.5	0.4	9.9	18.4	8.7	28.2	10	18	79.6
WAM06600- 009134	26	10	7	6.2	3.4	9.2	11.4	5.2	26	10	20	77.3