King County Freshwater Benthic Macroinvertebrate Sampling and Analysis Plan

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King County

Department of Natural Resources and Parks
Water and Land Resources Division
Science and Technical Support Section
King Street Center, KSC-NR-0704
201 South Jackson Street, Suite 704
Seattle, WA 98104
206-477-4800 TTY Relay: 711
www.kingcounty.gov/EnvironmentalScience

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Submitted by:

Kate Macneale, Liora Llewellyn, and Beth Sosik
King County Water and Land Resources Division
Department of Natural Resources and Parks

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1.0 INTRODUCTION

This Sampling and Analysis Plan (SAP) presents information about King County's Ambient Freshwater Benthic Macroinvertebrate Monitoring Program ("program") and describes the sampling and analysis methods. The primary objective of the program is to characterize the status and trends in the biological condition of stream sites across King County, WA. Stream biological condition is assessed using the Puget Lowlands Benthic Index of Biotic Integrity (B-IBI), which scores sites based on the diversity and relative abundance of benthic macroinvertebrates present at each site. Habitat information is also collected at each site to help characterize local conditions and better understand the influence of habitat conditions on B-IBI scores.

This SAP replaces the previous SAP (*Greater Lake Washington and Green-Duwamish River Watersheds Wadeable Freshwater Streams Benthic Macroinvertebrate Sampling and Analysis Plan* [2002]). Changes to the 2002 SAP have been implemented over time, as described in Appendix A.

1.1 History of the Program

There is a long history of using benthic macroinvertebrate data to assess and characterize stream condition and water quality in King County. King County staff first collected macroinvertebrate samples in 1976, to establish baseline stream health information on a majority of streams in the Lake Washington and Green River basins (White and Martin 1978). The sample analysis at the time was rudimentary, with insects identified only to family, but the methods and results used laid the foundation for more comprehensive monitoring projects and programs.

In the 1990s, King County Water and Land Resources Division (WLRD) initiated several benthic macroinvertebrate monitoring projects to address site-specific or project-related questions. The Basin Management Evaluation Program (BMEP) was initiated in the Soos Creek basin in 1994 and included benthic macroinvertebrate monitoring. This project was expanded in 1995 to include sampling throughout six basins. Additional macroinvertebrate monitoring projects included monitoring streams in forested watersheds receiving application of biosolids and a monitoring in the Trilogy and Redmond Ridge Urban Planned Developments to assess effectiveness of stormwater treatment facilities.

While these efforts helped inform specific monitoring and management goals, WLRD staff recognized a more comprehensive sampling design was needed to assess stream condition status and trends throughout the county. Thus, in 2002 and 2003, a stratified random design was used to select 163 sampling sites among 20 subbasins in the Green-Duwamish and Greater Lake Washington watersheds (portions of Water Resource Inventory Areas [WRIAs] 8 and 9; King County 2004). Most of these sites had not been previously monitored; however, some existing sites (e.g., some BEMP sites) that fell within the selected random grids were incorporated into the new ambient monitoring program. The macroinvertebrate monitoring program is part of the county's ongoing ambient water quality monitoring program which is funded through the Wastewater Treatment Division (WTD), and therefore the sampling area was limited to streams within the WTD service area. WLRD staff continue to sample most of the sites established in 2002 and 2003, although some sites have been dropped from the program due to lack of access or unsuitable habitat conditions (Appendix A). In addition, no samples were collected in 2004 due to funding limitations.

In 2014, Surface Water Management funds were designated to increase coverage of the sampling area outside of the WTD service area. Between 2014 and 2017, three sites in Boise Creek in the White River watershed (WRIA 10), and 39 sites in several subbasins within the King County portion of the Snoqualmie River watershed (WRIA 7) were added to the monitoring program.

A list King County's current ambient freshwater benthic macroinvertebrate monitoring program sampling locations is included in Appendix B.

1.2 Freshwater Benthic Macroinvertebrates

Benthic macroinvertebrates are small, bottom-dwelling animals visible to the naked eye and lacking backbones. Examples include insects, crustaceans, worms, snails, and clams that live in or on the streambed ("benthos") and other substrate within the stream. Healthy streams can support over 60 taxa and more than 5,000 individuals per square meter. Macroinvertebrates serve many important roles in aquatic ecosystems, but they are best known in the Pacific Northwest as the preferred prey of juvenile salmonids (Karr and Chu 1999). Since most taxa are relatively sedentary and require a year or more to develop, the diversity and relative abundance of macroinvertebrates at a site reflects local habitat conditions when the sample was taken, as well as conditions experienced throughout the year (Booth et al. 2001).

Benthic macroinvertebrate communities are effective biological indicators of stream condition because they reflect the cumulative impacts of multiple stressors in a watershed (e.g., Walsh 2006). Indices based on macroinvertebrate taxa assemblages are widely applied for assessment and management of aquatic resources since they can assist in determining linkages between observed ecological effects and environmental stressors. Because macroinvertebrate assemblages are good indicators of the biological health of stream systems, agencies and organizations throughout the world have adopted benthic macroinvertebrate monitoring programs. Although taxonomic experts are needed to identify and enumerate macroinvertebrates, sample collection is relatively inexpensive and requires minimal training.

1.3 Benthic Index of Biotic Integrity (B-IBI)

The Puget Lowlands Benthic Index of Biotic Integrity (B-IBI) is the primary data analysis tool used for this program. The B-IBI was developed specifically for Puget Sound Lowland streams as an integrative measure of stream biological health (Kleindl 1995, Fore et al. 1996, Karr 1998, Karr and Chu 1999, Fore et al. 2001, Morley and Karr 2002). The B-IBI is composed of ten metrics that characterize different aspects of the macroinvertebrate community, including taxonomic richness, tolerance and intolerance to environmental stressors, feeding ecology, and community structure (Table 1).

Table 1. Ten metrics that make up the Puget Lowlands Benthic Index of Biotic Integrity (B-IBI)

Metric	Description
Taxa richness	The number of unique taxa found in a sample. Overall taxa richness declines with increased urbanization.
Ephemeroptera richness	The number of unique mayfly taxa in a sample. Many mayfly taxa are intolerant of stressors associated with increased urbanization, and several are especially sensitive to fine sediment and contaminants.
Plecoptera richness	The number of unique stonefly taxa in a sample. Many stonefly taxa are intolerant of stressors associated with increased urbanization, including low dissolved oxygen concentrations and a lack of riparian vegetation.
Trichoptera richness	The number of unique caddisfly taxa in a sample. Several caddisfly taxa are relatively tolerant of environmental stressors, but generally taxa richness declines with increased fine sediment, loss of complex habitat and disruption of the stream food web.
Clinger richness	The number of taxa identified as clingers in a sample. Clingers have behavioral or morphological adaptations that allow them to attach and persist in stream riffles or other high energy habitats. These taxa tend to disappear when exposed to an excess of fine sediments.
Long-lived richness	The number of taxa in a sample that require more than a year to complete their life cycle. The number of these taxa decline if conditions vary year to year due to disturbances such as flooding or drought.
Intolerant richness	The number of especially sensitive taxa in a sample. These taxa are the first to disappear from a stream when urbanization in the watershed increases. These taxa represent approximately 15% of common taxa in the Puget Sound Lowlands.
Percent dominant	The percent of a sample composed of the three most abundant taxa. As urbanization increases in a watershed, sensitive taxa disappear and the relative abundance of a few tolerant taxa often increases.
Percent predator	The percent of a sample composed of individuals that are obligate predators. The structure of the stream food web changes with increased urbanization, often resulting in the loss of predators.
Tolerant percent	The percent of a sample composed of tolerant individuals. Tolerant taxa are defined as taxa that are more likely to be found in sites with greater watershed urbanization. These taxa represent approximately 15% of common taxa in the Puget Sound Lowlands.

The scoring process is described on the Puget Sound Stream Benthos (PSSB) web site (http://pugetsoundstreambenthos.org), but in summary, it is a sum of individual metric scores. Each metric describes a community characteristic that responds to human disturbance. Scores for each metric are calculated using equations derived from relationships between human disturbance and metric values (King County 2014a). For the B-IBI, the gradient of human disturbance used to derive the equations is urbanization, and specifically the percent of a watershed upstream of a site that is classified as urban (King

County 2014a). For eight of the metrics, values decrease with increasing urbanization and scores range from a low of 0 and a high of 10. For two metrics, percent dominant and tolerant percent, the relationship is reversed, and values increase with increasing urbanization. Thus, low values have a high score (10) and high values get a low score (0). The overall B-IBI score is the sum of the ten metric scores, which is used to categorize stream condition as excellent, good, fair, poor, or very poor (Table 2).

In 2014, the Puget Lowlands B-IBI was updated and recalibrated to incorporate new information and the significant amount of regional data that had been added to the Puget Sound Stream Benthos (PSSB; King County 2014a). The updated B-IBI is currently used to calculate B-IBI scores. Details about the recalibration process are available on the PSSB website at: http://pugetsoundstreambenthos.org/Projects/BIBI-Recalibration-Documentation.aspx).

Table 2. Benthic Index of Biotic Integrity scores and condition. Closed brackets [ ] include endpoints; open brackets ( ) exclude endpoints.

Condition of Biological Integrity	Description	B-IBI Score
Excellent	Comparable to least disturbed reference condition; overall high taxa diversity, particularly of Ephemeroptera (mayfly), Plecoptera (stonefly), Trichoptera (caddisfly), long-lived, clinger, and intolerant taxa; relative abundance of predators high.	[80-100]
Good	Slightly divergent from least disturbed condition; absence of some long-lived and intolerant taxa; slight decline in richness of Ephemeroptera, Plecoptera, and Trichoptera; proportion of tolerant individuals increases.	[60-80)
Fair	Total taxa richness reduced – particularly intolerant, long-lived, Ephemeroptera, Plecoptera, and clinger taxa; relative abundance of predators declines; proportion of tolerant individuals continues to increase.	[40-60)
Poor	Overall taxa diversity depressed; proportion of predators greatly reduced as is long-lived taxa richness; few Plecoptera or intolerant taxa present; dominance by three most abundant taxa often very high.	[20-40)
Very Poor	Overall taxa diversity very low and dominated by a few highly tolerant taxa; Ephemeroptera, Plecoptera, Trichoptera, clinger, long-lived, and intolerant taxa largely absent; relative abundance of predators very low.	[0-20)

2.0 PROGRAM DESIGN

2.1 Program Objective

The objective of the program is to use aquatic macroinvertebrate community data to characterize stream conditions within King County watersheds. Aquatic macroinvertebrate monitoring data are used to address the following questions:

- What is the current condition of streams within King County?
- Are conditions improving or declining over time?

2.2 Monitoring Program Sampling Area

The monitoring program sampling area includes wadeable streams in basins within King County, as well as several basins in Snohomish County that drain to streams within King County (Figure 1). The study area is within four WRIAs (Table 3, Figure 1). Sites sampled as part of the Ambient Monitoring Program were selected randomly (Table 3), whereas sites sampled for other projects have been targeted. Data from these targeted sites are also used to characterize ambient conditions in King County streams, but are identified as separate projects (Table 3). Project names listed in Table 3 are the same as those listed in the PSSB.

Table 3. Projects/Programs included in the King County Ambient Freshwater Benthic Macroinvertebrate Monitoring Program, and the number of sites scheduled for sampling in 2020.

Project/Program	Method used for Site Selection	WRIA	Watershed	Number of Sites
Ambient Monitoring Program	Stratified random design using Master Sample	7	Snohomish/Snoqualmie	39
	Stratified random design using 0.33 mile^2 grid	8	Greater Lake Washington	78
	Stratified random design using 0.33 mile^2 grid	9	Green/Duwamish	44
Boise Creek Ambient	Targeted	10	Puyallup/White	3
Vashon	Targeted	9	Green/Duwamish	3

As indicated in Section 1.1, some sites have been dropped due to lack of access or significant habitat changes causing the sites to be unsuitable for the riffle-based sampling protocol used by this program. Since 2002, 21 sites in WRIA 8 and 20 sites in WRIA 9 have been dropped. In the future, dropped sites may be reexamined to determine if access or habitat conditions have changed or new sites may be identified to ensure the subbasin can be sufficiently characterized.

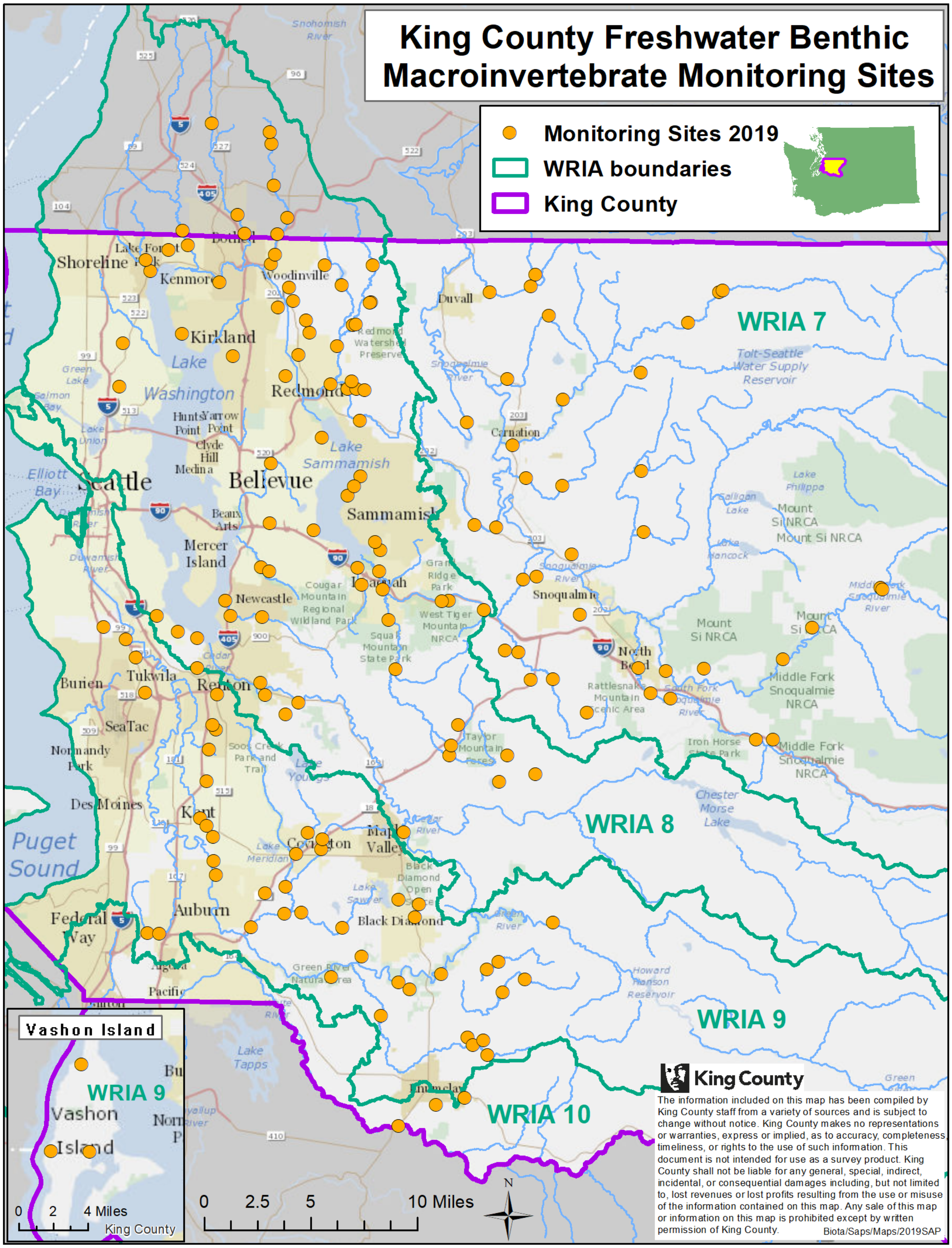

Figure 1. Freshwater benthic macroinvertebrate monitoring sites as of 2019.

2.2.1 Snohomish/Snoqualmie Watershed (WRIA 7)

As of 2020, samples are collected annually from 39 locations within the King County portion of the Snohomish/Snoqualmie Watershed (WRIA 7). This portion of the watershed includes the Snoqualmie River and its tributaries (i.e., Raging and Tolt rivers and their tributaries). The headwaters originate in the Cascade Mountains and discharge into Puget Sound near the City of Everett. Land use in the Snoqualmie basin includes active forestry, agriculture, and increasing urban and suburban expansion. The Tolt River (South Fork) provides a third of the City of Seattle's drinking watery. The WRIA 7 sites were phased into the Ambient Program starting in 2014.

2.2.2 Greater Lake Washington Watershed (WRIA 8)

As of 2020, samples are collected from 78 locations in the Greater Lake Washington Watershed (WRIA 8). This watershed has a drainage area of approximately 692 square miles, and includes the Cedar and Sammamish Rivers, lakes Washington and Sammamish and a number of smaller creeks that ultimately drain into Puget Sound. WRIA 8 is the most densely populated watershed in the state and includes two of the region's most urban areas including Bellevue and portions of Seattle.

2.2.3 Green/Duwamish Watershed (WRIA 9)

As of 2020, samples are collected from 47 locations in the Green/Duwamish Watershed (WRIA 9); this total includes three sites on Vashon Island. This watershed originates on the slopes of Mount Rainier, drains to Elliot Bay, and includes several large creeks (e.g., Soos and Newaukum) as well as the Green River, which becomes the Duwamish River downstream of its confluence with the Black River. The watershed drainage area is approximately 492 square miles. Terrain and land use vary from forested headwater areas at the crest of the Cascade Mountains to industrial facilities in the Duwamish estuary. The basin includes the Howard Hanson dam, portions of Seattle, and the varied agricultural, residential, commercial, and industrial land uses in the Green River Valley and Enumclaw plateau. For the purposes of salmon conservation planning (and this monitoring program), Vashon-Maury Island (WRIA 15) has been included in WRIA 9.

2.2.4 Puyallup/White Watershed (WRIA 10)

A small portion of the Puyallup/White River Watershed (WRIA 10) is within King County (Figure 1). This watershed originates from the glaciers of Mount Rainier, and includes the Carbon, White, and Puyallup rivers that flow through the City of Tacoma and discharge to Puget Sound.

In 2014, staff established annual macroinvertebrate sampling sites at three sites in the Boise Creek watershed that had previously been monitored for water quality. Boise Creek originates near the City of Enumclaw and flows into the White River. Land use in the Boise Creek basin includes forest management in the upper watershed and agriculture and residential and commercial development in the lower portion.

2.3 Site Selection

Ambient Monitoring Program sites were randomly selected in one of two ways. The first selection process was initiated in 2002 to select sites within WRIAs 8 and 9, and the second was used in 2014 to select sites in WRIA 7. Although the methods vary slightly, both result in a probabilistic random sampling design that can be used to characterize conditions across streams in the sampled subbasins.

For sites identified in 2002, WLRD staff used the County's Geographic Information System (GIS) to overlay a 0.33 square mile (0.85 square kilometer) grid on a map of the Green-Duwamish and Greater Lake Washington watersheds (King County 2004). All grids that included stream segments were assigned a random number and grouped by subbasin. Sites were visited in order and each was evaluated to determine if it was accessible and had suitable habitat. Site suitability was defined as a stream reach with at least one riffle, a minimum channel width of one foot, and a water depth ranging from one inch to one foot during low-flow summer conditions. WLRD staff evaluated sites on private property only after permission was granted from a landowner. Inaccessible or unsuitable sites were eliminated and the next potential site on the list was evaluated. This process was followed until up to 10 sites in each subbasin had been identified.

In 2014, WLRD staff used the Washington Master Sample to identify potential sampling locations in several WRIA 7 subbasins. The Master Sample is a state-wide list of 1-km stream reaches that were assigned a random number using a Generalized Random Tessellation Stratified (GRTS) design (Stevens and Olsen 2004, Olsen et al. 2012, Ecology 2008). WLRD staff stratified the list by subbasin and limited potential sites to first through fourth Strahler order streams, at or below 500 m elevation. Subbasins included the Patterson, Cherry, Tokul, Harris, Stossel, Griffin, Tolt, Raging, and the Middle and South Forks of the Snoqualmie. The number of target sites per subbasin was determined by the size of the subbasin, with the target number ranging from 6 in largest subbasin to 1 in the smallest subbasin. Suitability was evaluated in a similar method used in 2002, with an additional criterion that each site had to be within a one-mile hike from a road. As before, WLRD staff evaluated sites on private property only after permission was granted from a landowner.

3.0 SITE VISIT AND SAMPLE COLLECTION METHODS

King County's protocol for sampling benthic macroinvertebrates in wadeable streams follows recognized methods used by others in the region (e.g., Karr and Chu 1999, Plotnikoff and Wiseman 2001, Hayslip 2007, Ecology 2016a) with some modifications. King County currently follows a riffle-based collection method, collecting 8, 1-ft^2 samples from a stream reach and compositing those into a single sample. Staff also collect habitat data associated with the sampling site.

The sections below describe activities that occur before sampling (e.g., training, confirming site access), as well as the detailed protocols for collecting macroinvertebrate samples and the associated habitat data. Protocols have changed since this program was initiated in 2002; changes are summarized in Appendix A.

3.1 Personnel and Training

Sampling is conducted by teams of at least two trained staff. Prior to sampling, all staff review protocols and participate in an annual field training event. The lead staff scientist demonstrates proper use of field equipment and reviews habitat definitions and data collection protocols to ensure consistent sampling across personnel. The lead scientist ensures all staff have also completed appropriate safely training prior to field sampling. To ensure consistency in sampling protocols across field crews, a staff member with significant prior sampling experience accompanies the field crew during the first week of sampling each season. Staff rotate among teams throughout the season to help ensure all staff are following protocols consistently.

3.2 Site Access

Staff must obtain permission to access sites on private property before sampling. If site access requires crossing multiple property boundaries, permission must be obtained from all affected landowners. If access is denied or the landowner cannot be contacted within the sampling season, the site may be shifted up- or downstream within 200 meters from its original location, assuming there are no significant changes in flow due to the shift (e.g., no tributary confluence). Staff must record how permission was obtained for each site, as well as any location changes.

3.3 Sample Timing

Macroinvertebrate samples are collected during low-flow conditions between mid-July and early October. This period provides adequate time for stream communities to stabilize following disturbances such as high winter and spring flows, and ensures the presence of larger macroinvertebrates for easier identification.

To ensure staff safety and minimize any immediate effect of high flow conditions on B-IBI scores, samples are not collected during and immediately following heavy rain fall (>0.5 inches within 24 hours). Sampling is also delayed as late as possible in the sampling season at sites downstream of recent construction or major landscape changes.

It is assumed that the B-IBI score reflects conditions regardless of when the sample is collected during the designated sampling window, and is not sensitive to changes in species composition that may occur naturally over this long sampling period.

3.4 Sample Equipment

The following gear is required at each sampling site. It is recommended that staff bring duplicates of items that are easily lost or broken (e.g., pencils, labels, forceps, ping-pong balls). All sampling equipment and wading gear must be clean of organisms, and if needed, decontaminated before use (Section 3.8).

- Surber sampler (1-ft^2 frame with 500-μm mesh net, and removable plankton cup with 500-μm mesh openings); alternatively, for sites with swift current and large cobble, use a D-Frame kicknet with 500-μm mesh net and a frame 1 ft wide and 1 ft tall.
- Sieve with 500-μm screen
- Ruler (at least 30 cm), with carabiner
- Garden weed tool, marked 10 cm from tip
- Denatured ethyl alcohol, 95% or greater (plan for at least 1 L per sample)
- Sample containers with lids (at least 1, 2-liter and 1, 1-liter container)
- Clipboard
- Field datasheet on waterproof paper (or electronic form and device)
- Camera
- Pencil
- Permanent marker
- Stickers for labeling exterior of sample bottle(s)
- Waterproof labels to be placed inside sample bottle(s)
- Garden sprayer (hand pump style)
- Tools for transferring sample from dishpan to bottle (spatula, slotted spoon, plastic spoon, fine-tip forceps)
- Dishpan, white or light-colored
- Metric measuring tape (at least 30 m)
- Laser range finder
- Stopwatch
- Ping-pong ball

- Convex densiometer
- Wading gear
- WA State Scientific Collection permit

3.5 Reach Selection

Samples are collected from riffles, or if riffles are not present, from reaches with fast-moving water. Riffles are characterized by relatively fast moving, broken water over large gravel or cobble substrate, and are common features of wadeable streams in the Pacific Northwest. King County targets riffles because the Puget Lowland B-IBI was developed for samples collected from riffles (Fore et al. 1996, Karr 1998) and previous collection protocols targeted riffles (King County 2002).

Samples are intended to characterize the communities found in fast-flowing habitats across the stream reach and are therefore are collected from multiple, distinct riffles. Staff identify riffles prior to sampling, and care must be taken to avoid walking in the stream or causing any disturbance upstream of the areas that will be sampled.

Ideally, staff collect two, 1-ft^2 samples from each of four riffles (total of 8 Surber net collections) and then composite them into a single sample (Figure 2). If possible, samples are collected upstream of a road crossing, but for consistency this may be amended based on the location of the previous year's collection site. The sample reach is the length of stream containing the sampled riffles, measured in meters. In Figure 2, this is the distance from downstream end of riffle 1 to the upstream end of riffle 4.

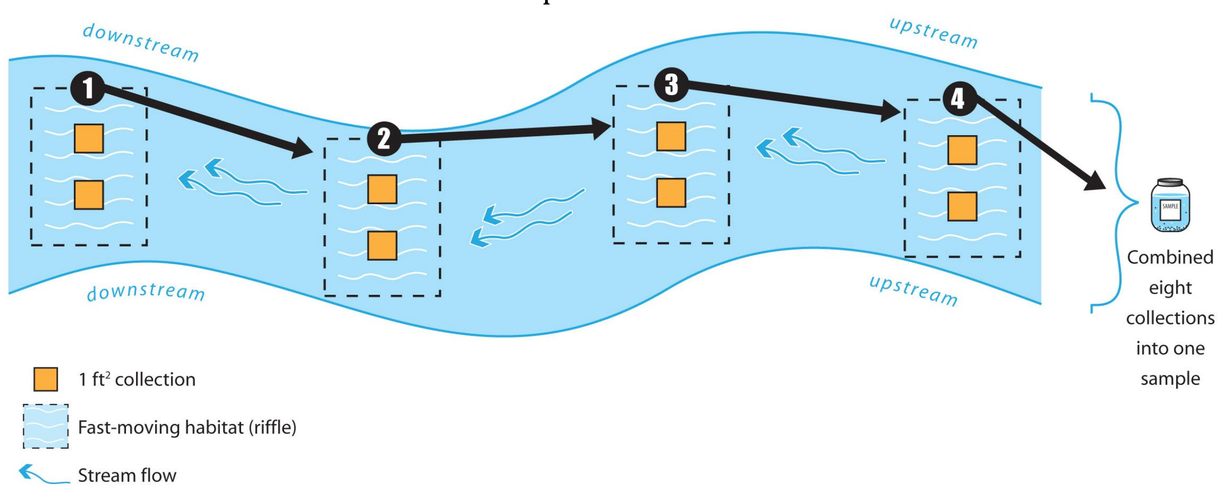

Figure 2. Stream reach for benthic macroinvertebrate sample collection: 8 ft^2, ideally from 4 riffles, 2 collections from each riffle, composited into a single sample, moving from downstream (1) to upstream (4).

If there are not four riffles within the accessible reach, staff may collect 8 samples from fewer riffles. If there are no well-defined riffles present, staff may collect samples from the fastest flowing, most turbulent, and non-depositional locations. At a minimum, there must be sufficient water depth and flow to move the dislodged organisms over the lip of the

Surber frame and into the net. Riffles split around a bar or island are considered to be a single riffle if they continue/meet above or below the obstruction.

3.6 Sample Collection

Once four riffles are identified, staff should begin sample collection at the downstream-most riffle to prevent disturbance of subsequent sampling areas. The Surber net is placed in a representative location within a riffle; this includes the thalweg (deepest and often fastest-flowing section of the riffle) as well as adjacent areas that may not be as fast or deep (Figure 2). Combined, the 8 samples should reflect the variety of riffle habitats present in the sampled stream reach.

To collect a sample, the Surber net is placed firmly onto the substrate with the net opening facing upstream. The net frame is then pressed down and settled into the substrate, creating a seal against the substrate to prevent organisms from washing under the frame rather than into the net. If large cobble or other material lying under the edge of the frame prevents a good seal, the material should be immediately pulled up within the perimeter of the frame (if <25% of a piece of large cobble is in the way, move it outside of the frame). Water depth must be at least several centimeters deep (to wash invertebrates into net), and no more than 1 ft deep (to prevent organisms from being flush up and out of net). In faster flowing streams, or those in which riffles are consistently deeper, staff may use a D-net rather than a Surber net to collect the sample.

Once the net is placed, staff quickly collect the sample to minimize movement of organisms into or out of the 1-ft sampling area. When samples are taken in shallow water, any rocks or debris downstream of the frame (and underneath the net) potentially impeding the flow of water and invertebrates into the net and sample cup should be removed. Care must be taken to not disturb the substrate upstream and outside of the frame before or when the net is in place.

All large objects (e.g., large cobble and woody debris) within the Surber frame should be picked up and scrubbed by hand while submerged. Once all organisms are removed, the object is discarded downstream of the sample collection area. Organic matter (sticks, algae, and leaves) should stay within the sample area and should be included in the sample. Aquatic vegetation growing on the substrate should also be scrubbed off and included in the sample. When the substrate is too large to move, but less than a quarter of the sample area, scrub the surface within the sample area. This entire process should only take a minute or less.

Staff then use a garden weed tool to vigorously agitate the substrate within the sample frame to a depth of approximately 10 cm for 60 seconds. The frame must stay securely anchored to the substrate during this process. After 60 seconds of agitation, the lip of the Surber frame should be lifted above the flow so that no additional organisms are collected. If there are concerns that glass or other sharp objects may be present in the benthos, staff are advised not to wash rocks and debris by hand and only agitate the substrate with the weed tool.

Staff should inspect the sample for vertebrates, mussels, and crayfish. If any are found, staff should record their presence on the field sheet. If salmonids, sculpin, and mussels are collected, staff must record the number and status (alive or dead) and return them to the stream as quickly as possible. Dead fish and all mussels should be photographed. Crayfish collected in the sample are counted, recorded, and retained.

During sampling, rocks or sticks may wash into the net. Any materials too large to fit through the cod end/sample cup neck should be scrubbed and inspected inside the net. Once all organisms are removed, large rocks may be discarded in the stream. Smaller rocks and organic debris should be kept and placed in the sample container.

If the sample cup fills and overflows into the net before all 8 samples have been collected, the cup's contents may be poured into a clean dishpan and the cup reattached to complete sampling. Once the collection is complete, staff should rinse their hands and the garden tool with filtered stream water to ensure any attached organisms are washed into the sample net.

3.7 Sample Processing and Preservation

All sample material is transferred to a clean plastic tub, and then transferred to one or more sample bottles. Any invertebrates attached to the inside of the net and cup are rinsed with filtered stream water (sieved through 500-μm mesh) into the tub. Rocks larger than 3 cm in diameter and sticks can be rinsed and discarded once all attached invertebrates have been removed, but all other material is retained. Excess water is drained from the sample through a 500-um sieve, and all material is transferred to one or more 1- or 2-L sample bottles. Sample material should fill no more than one-half of the bottle to allow sufficient room for ethanol. Finally, the sample net, cup, and tub should be carefully inspected, and any attached invertebrates removed with tweezers and placed in bottles.

Within an hour of collection, ethanol (100%, denatured) is added to each bottle so the sample material is entirely covered. The final storage solution must be at least 70% ethanol to prevent sample decay.

Sample bottles are labeled with both internal and external labels including the sample name, date, number of bottles per sample, and initials of the sampling crew. The sample name is the site code followed by an underscore and the last two digits of the year. For example, the sample from 08BEA3312 collected in 2019 was named "08BEA3312_19". If a replicate sample is collected, an "_R" is included at the end of the replicate sample name to distinguish the original and replicate samples (e.g., 08BEA3312_19_R).

3.7.1 Sample Storage

Collected samples are placed in a cooler for transport from the field and stored at room temperature in a secure storage room. Access is restricted to authorized staff. A chain-of-custody (COC) form is filled out each day samples are collected and kept with them at all times.

3.7.2 Chain of Custody

To maintain the legal integrity of all samples, a COC procedure is followed by all project staff. Prior to sampling, a blank COC form (Appendix D) is printed and placed in the sample storage area. If a correction is required on the COC form, a single line is drawn through the error so it remains legible, and the correction is written adjacent to the error as well as the author's initials and date.

When samples are ready for transport to the Taxonomic Laboratory, the completed COC form is scanned and saved electronically. The original COC is provided to the recipient when the samples are transferred and it accompanies the samples at all times.

3.8 Decontamination Procedures

Decontamination procedures are implemented to reduce the risk of spreading invasive species, noxious weeds, and pathogens between sites and basins. Of particular concern in King County is the invasive New Zealand mudsnail (*Potamopyrgus antipodarum;* NZMS). Like other invasive species, weeds, and pathogens, NZMS are small, difficult to kill, and easily spread if gear is not properly decontaminated. More information about NZMS and other non-native species is included in Sections 4.7 and 4.8.3.

King County's decontamination protocols are based on standards established by Ecology (Ecology 2016b) and the Washington Department of Fish and Wildlife (WDFW 2016). State standards require specific Level 1 and Level 2 decontamination procedures based on sampling locations and risk (see below). Schedules are also planned to ensure teams work within a single basin during a sampling day, and move from the upper to lower watershed whenever possible to reduce the risk of cross contamination within and between basins. Field staff decontaminate sampling equipment and gear between sites and at the end of each sampling day. In addition, King County staff do not wear felt-soled wading boots because these have been implicated in the spread of invasive species and pathogens.

3.8.1 Level 1 Decontamination

Level one, or "basic," decontamination protocols are required whenever moving from one water body to another, regardless of whether it is in the same basin. The steps of Level 1 decontamination include cleaning, draining, and rinsing all gear or equipment that came into contact with the water or stream bed. Each vehicle is equipped with a Level 1 kit that includes a tub containing a sturdy bristle scrub brush, a pressurized pump sprayer filled with clean water, and an extra container of rinse water. Waders, boots, and sampling equipment are scrubbed and rinsed after each site. Rinse water does not need to be contained for Level 1 decontamination.

3.8.2 Level 2 Decontamination

Level 2 or "high risk situations" decontamination protocols are required when moving across WRIA boundaries, when leaving known infested waters, and before entering protected or highly sensitive sites. Level 2 decontamination is designed to kill/eradicate invasive species, and is done after Level 1 procedures have been completed. Freezing gear and equipment is the preferred Level 2 decontamination procedure used by King County; if that is not possible, Virkon is used as an alternative.

All field equipment and wading gear that has been immersed in a stream or been in contact with the macroinvertebrate sample is placed in a chest freezer at a maximum of 14°F (-10°C) for at least 8 hours. The Washington Invasive Species Council recommends gear be held at -4°C for a minimum of 4 hours to achieve 100% NZMS mortality (time starts once the equipment reaches -4°C) (Ecology 2016b). Freezing has also been shown to be effective against the benthic diatom Didymo (*Didymosphenia geminate*), though it may not be effective against chytrid fungus (*Batrachochytrium dendrobatidis*) or the fish pathogen whirling disease (*Myxobolus cerebralis*) (Root and O'Reilly 2012, Kilroy et al. 2006, WDFW 2016).

As of May 2020, NZMSs have been found in many tributaries to Lake Washington, as well as some tributaries to Lake Sammamish. In addition, NZMSs have been found in streams draining directly to Puget Sound as well as the Green and White rivers. These streams include: Thornton, Pipers, McAleer, May, Mapes, Maplewood, Lyon, Sunset, Longfellow, Kelsey, Taylor, High School, Big Soos, North, Country, and Boise creeks. Staff should conduct Level 2 decontamination as standard practice when working in all of these basins.

4.0 HABITAT EVALUATION

Instream and riparian habitat conditions are assessed in conjunction with macroinvertebrate sample collection to document local conditions that may influence B-IBI scores. The habitat protocols described below are intended to be conducted rapidly (within approximately an hour) and therefore are not as comprehensive as habitat protocols used by others (e.g., Ecology or the US Forest Service). Most of the measurements and assessments can be made by one staff member, but for some measures a second staff member may be helpful. Staff record data on a waterproof paper field form (Appendix C), and, back in the office, enter data into an ArcGIS database. Staff also take photos at each site.

Continuous temperature is measured with data loggers at select sites. Protocols for deploying temperature loggers and checking data are included in the King County Continuous Temperature Monitoring SAP (2020).

Habitat collection methods have been revised since the program was initiated in 2002. Method changes are summarized in Appendix A.

4.1 Descriptive Site Information

Staff describe briefly the location and weather on the sampling day. Staff should note any changes to the location from previous years (e.g., site moved upstream 100 m because of blocked access point). Staff record the date and the time the survey starts, as well as names of staff collecting the macroinvertebrate sample and conducting the habitat survey. Staff record the current weather conditions (sunny, partly cloudy [10-50%], mostly cloudy [>50%], or raining), and recent weather conditions (rain events within past 7 days, as well as precipitation intensity). Water clarity is also observed and noted. Any additional relevant observations are recorded in the notes section.

4.2 Sample Reach Dimensions

Reach dimensions are recorded to help characterize the relative area of the sampled reach. Reach widths include wetted and bankfull widths, but reach length is somewhat arbitrary because it depends on the length of stream that staff have permission to access and the number of riffles within that area. Ideally, a reach will encompass four distinct riffles and be representative of the greater stream system. Reach length is measured in meters from the downstream edge of the first sampled riffle to the upstream edge of the last sampled riffle. A tape measure or range finder should be used to measure length along the thalweg to the nearest meter.

Reach length is divided into three equal areas spanning the sampled riffles (Figure 3). At each of the four cross sections spanning the reach, staff record the wetted width, bankfull width, bankfull depth, and thalweg depth (Figure 4). Each width measurement is recorded in meters to the nearest centimeter, and each depth measurement is recorded in centimeters to the nearest centimeter.

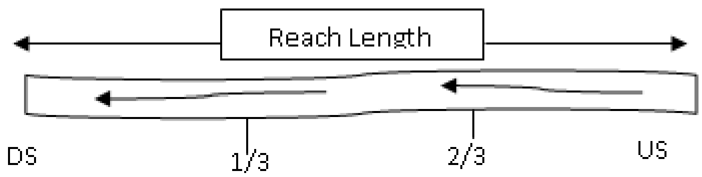

Figure 3. Reach length and four cross sections (at downstream [DS], 1/3, 2/3, and upstream [US]), indicating where wetted width, bankfull width, bankfull depth, and thalweg depth measurements are taken.

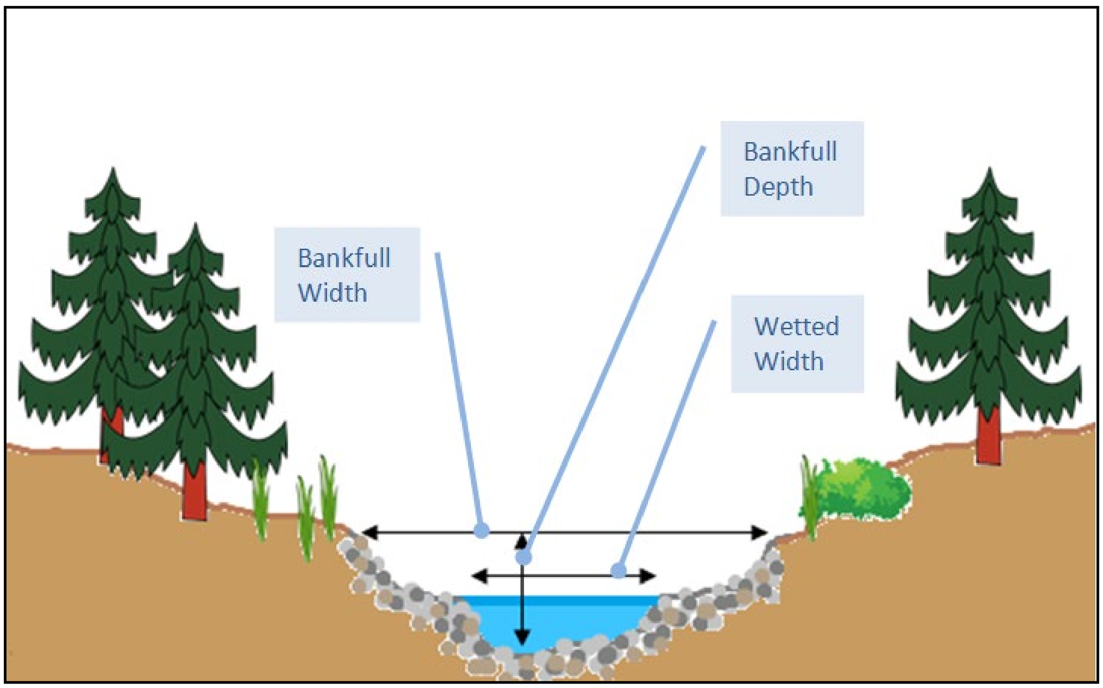

Figure 4. Instream features: bankfull width, wetted width, and bankfull depth.

Determination of both wetted and bankfull widths should be measured at approximately a right angle to the direction of flow. The wetted width is defined as the distance between the water's edge on one bank to the other and is measured at each of the four cross sections. If there is a dry gravel bar or island within the cross section, its width is subtracted from the wetted width. Bankfull width is measured at the point of incipient flooding. This point is indicated by sand or silt deposits at the active scour mark, a break in stream bank slope, perennial vegetation limit, or rock discoloration and root hair exposure, including any gravel bars and split channels or undercutting. In the rare instance of an island rising above the bankfull width, indicated by perennial vegetation, the width of the island above bankfull height is excluded from the bankfull width measurement. A laser range finder may be used for larger streams or rivers where the entire width is difficult to access.

The bankfull depth is measured at the thalweg from the substrate to the height of where the bankfull width was measured (Figure 4). The thalweg depth, measured at the thalweg from the substrate to the water surface, is also recorded at each of the four cross sections.

Staff measure and record an average riffle depth for each of the 8 sampled areas. Each average depth is approximated from multiple depths taken within the 1-ft^2 Surber sample frame. Riffle depths are measured before removal of substrate and recorded to the nearest centimeter.

4.3 Stream Bank Stability

Stream bank stability characterizes potential local sources of fine sediments. Stability of the right and left banks (determined while facing downstream) is assessed for the entire sampled reach and categorized using the US Environmental Protection Agency's (EPA) Rapid Bioassessment Protocols (Barbour et. al. 1999) (Table 4).

Table 4. Bank stability classification based on the EPA's Rapid Bioassessment Protocols.

BANK STABILITY *(circle one for each bank throughout reach, bank side facing downstream)*	**Bank stable** Evidence of erosion or bank failure absent or minimal; little potential for future problems. <5% of bank affected.		**Moderately stable** Infrequent, small areas of erosion mostly healed over. 5-30% of bank in reach has areas of erosion.			**Moderately unstable** 30-60% of bank in reach has areas of erosion; high erosion potential during floods.			**Unstable** Many eroded areas; "raw" areas frequent along straight sections and bends; obvious bank sloughing; 60-100% of bank has erosional scars.		
Right Bank	**10**	**9**	**8**	**7**	**6**	**5**	**4**	**3**	**2**	**1**	**0**
Left Bank	**10**	**9**	**8**	**7**	**6**	**5**	**4**	**3**	**2**	**1**	**0**

4.4 Velocity

Surface flow velocity of riffle habitats is measured using the float method (Ecology 2017a). Velocity is estimated by measuring the time a ping-pong ball travels over a known distance in a representative riffle (following sample collection). Ideally the travel distance (measured in meters to the nearest cm) should be at least three channel widths long; if this is not feasible, the travel distance should be as long as possible. The ping-pong ball is released upstream of the starting point so that it is traveling at the speed of the current when it passes the top of the measured reach. Field staff should make sure they are standing upstream of the measured reach to ensure that they are not influencing the flow. If the ball becomes stranded in an eddy or interrupted by rocks or debris in the path, the measurement should be restarted. When the ball passes the endpoint, the stopwatch is stopped and the time is recorded. The measurement is repeated three times.

4.5 Large Woody Debris and Pools

Staff count the number of pools and pieces of large woody debris (LWD) present within the reach to characterize habitat complexity. For this protocol, LWD is define as dead wood, ≥2 m long with some portion ≥10 cm in diameter. LWD must lie, at least partially, within the bankfull width of the stream to be counted. Rootwads that are ≥0.03 m^3 (equal to or larger that the Surber 1-ft^3 frame) are also counted as LWD. Logs spanning the channel above bankfull are not included in counts of LWD. If a log or rootwad is partially buried but appears to meet the size criteria, it is counted as LWD.

Pools are defined as relatively deep, depositional areas with reduced velocity. For this protocol, pools are counted only if the deepest point (max depth) is ≥1.5x the crest depth (Figure 5) and at least as long as the width of the wetted channel. The crest depth is the shallowest thalweg depth within a pool, typically where the habitat is transitioning from a pool to a riffle or glide (Figure 5).

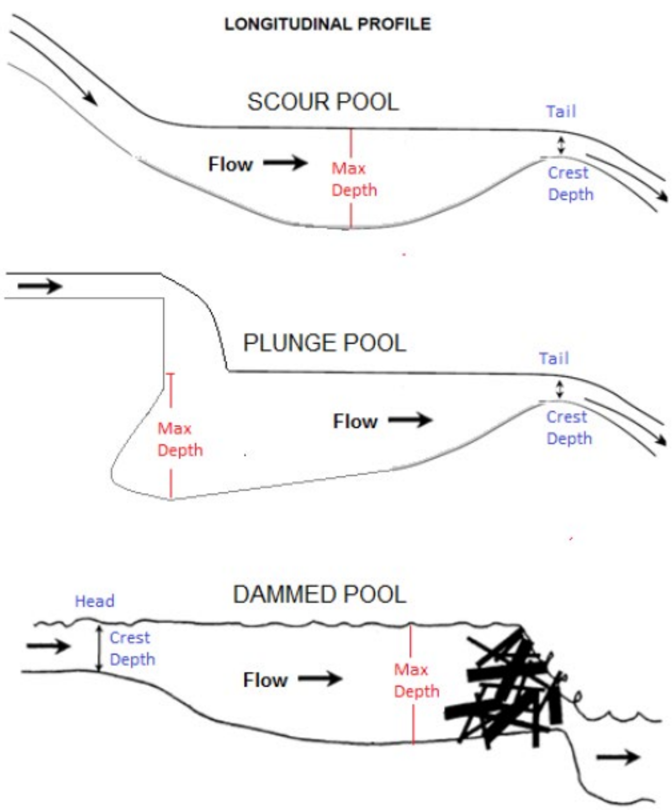

Figure 5. Examples of pool dimensions, including max and crest depths. Figure copied from Ecology (2017c).

4.6 Pebble Count

Pebble counts help characterize the size distribution of the benthic substrate in riffles. Pebble counts were first described by Wolman (1954), and King County follows a modified Wolman Count method. Staff establish a transect spanning one of the sampled riffles within the middle of the reach. Starting at the wetted edge, and then at regular intervals along the transect spanning the riffle, staff measure the size of a randomly selected substrate particle using the gravelometer (Figure 6). The spacing interval can vary by stream, but should not vary along the transect (e.g., if the interval is 20 cm between measuring points, that distance should consistent across the transect). The interval should be wider than the average substrate size so the same pebble/boulder is not measured multiple times. Each particle is binned by size class (Table 5) according to the smallest area it fits through in the gravelometer (Figure 6).

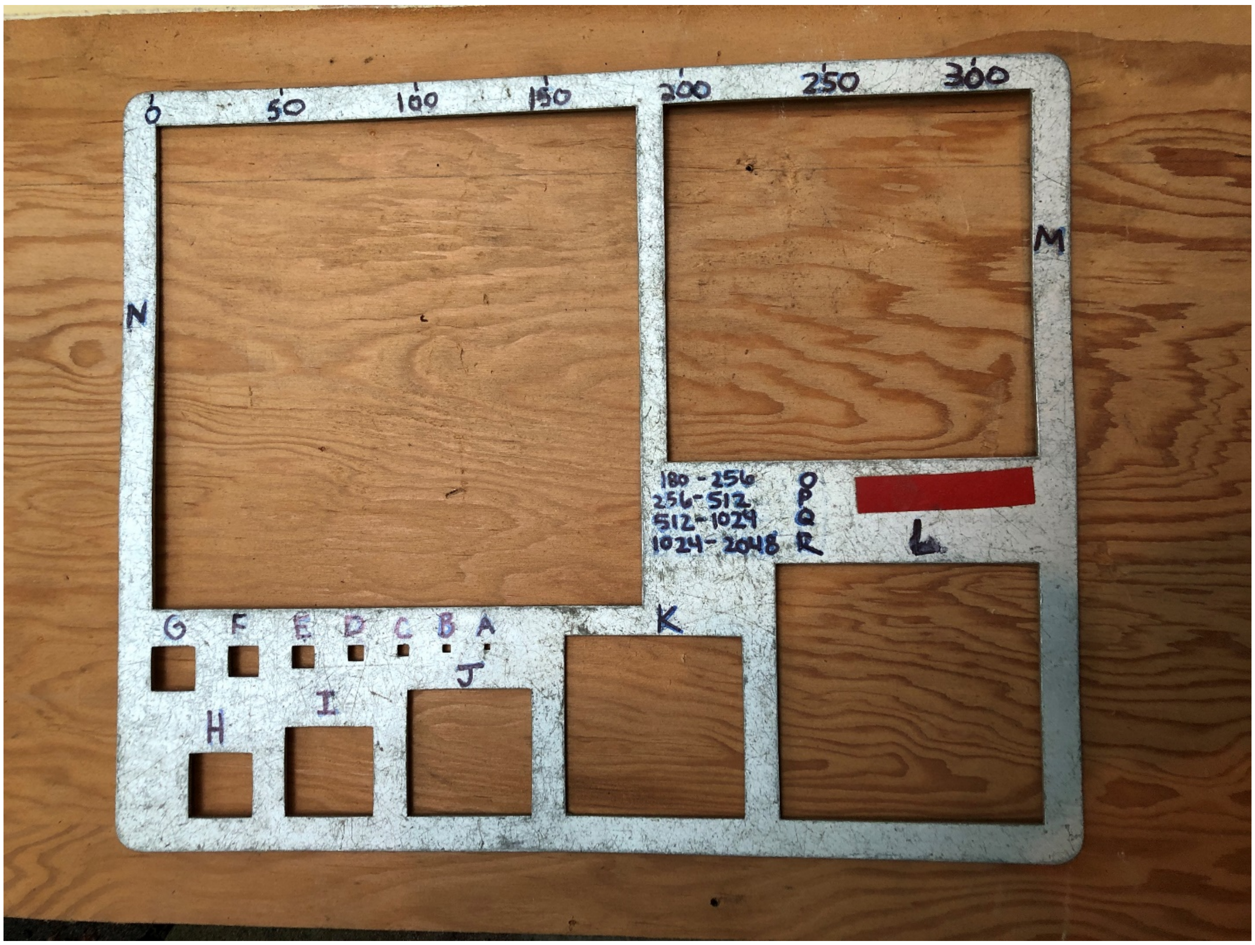

Figure 6. Gravelometer used to measure substrate size. A particle is given the letter code for the smallest opening it fits through.

Table 5. Substrate size classes for measuring the smallest width of particles for the Wolman pebble count.

Substrate Class	Size Class (mm)	Letter Code
Sand	0 - 2	A
Very Fine Gravel	>2 - 2.8	B
Very Fine Gravel	>2.8 - 4	C
Fine Gravel	>4 - 5.6	D
Fine Gravel	>5.6 - 8	E
Medium Gravel	>8 - 11	F

Substrate Class	Size Class (mm)	Letter Code
Medium Gravel	>11 - 16	G
Coarse Gravel	>16 - 22.6	H
Coarse Gravel	>22.6 - 32	I
Very Coarse Gravel	>32 - 45	J
Very Coarse Gravel	>45 - 64	K
Small Cobble	>64 - 90	L
Medium Cobble	>90 - 128	M
Large Cobble	>128 - 180	N
Very Large Cobble	>180 - 256	O
Small Boulder	>256 - 512	P
Medium Boulder	>512 - 1024	Q
Very Large Boulder	>1024 - 2048	R

Staff repeat these measurements, across as many transects as necessary, until at least 100 particles are measured. Once a new transect is initiated, staff must complete the process over the entire length even if the 100 particle quota is reached before the end of the transect.

For embedded particles or those too large to move, measure the shortest axis visible. Bedrock, garbage, construction debris, or organic materials should not be considered in the pebble count. If the crew member encounters fine sediment completely covering a rock (not sporadically), the fine material is measured rather than the rock.

4.7 Aquatic Invasive Species

As discussed in Section 3.8 above, staff decontaminate gear in part to reduce risk of spreading invasive species among sites. Of primary concern in King County streams are NZMS. These aquatic snails are tiny (3-6 mm in length), thrive in disturbed watersheds, and are parthenogenetic (capable of asexual reproduction). They can survive passage through the guts of fish and be transported alive, quickly reproducing to densities of more than 100,000 per m^2 (WDFW 2020, Kerans et al. 2005, King County 2017). NZMS have no predators and compete with native snails and aquatic insects for food resources.

At each site, staff inspect five rocks in two habitats (riffles, runs, and/or pools) and note if NZMS are present. If NZMSs, or snails that are likely NZMSs, are found in a basin where they have not previously been detected, staff will collect several individuals to confirm taxonomic identification. The specimens are sent to the taxonomic lab and/or an independent taxonomist to verify the identification.

Staff are also instructed to look for African clawed frog (*Xenopus laevis*) in North Creek in the Sammamish River basin. If present, staff should report the finding to the Washington Invasive Species Council (https://invasivespecies.wa.gov/report-a-sighting/). Decontamination Level 1 procedures (Section 3.8) should prevent the spread of invasive frogs, but may not address potential pathogens carried by these frogs.

4.8 Riparian Habitat Assessment

Riparian habitat includes the physical attributes of the land area immediately adjacent to the stream. The following sections describe the habitat components included in the assessment. Changes to habitat assessment protocols since the program was initiated in 2002 are documented in Appendix A.

4.8.1 Densiometer

Riparian canopy cover can influence stream temperature, leaf litter and wood availability, bank stability, sediment delivery, and habitat quality for the terrestrial life stages of aquatic insects. Canopy cover is estimated using a Strickler-modified convex densiometer as per Ecology's Standard Operating Procedures (Ecology 2017b; Strickler 1959). The densiometer is modified by applying electrical or duct tape to the lower portion of the mirror, to ensure peripheral areas are not oversampled when taking four directional measurements.

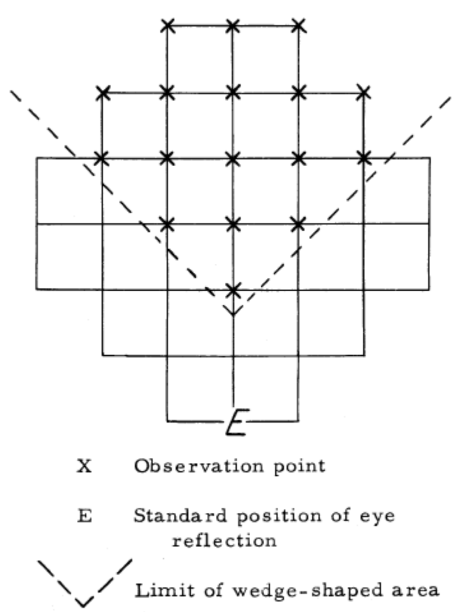

Figure 7. Diagram of the modified densiometer mirror, with 17 observation points (x). Figure copied from Strickler (1959).

Measurements are taken at three locations within the sample reach: the downstream end of the reach, middle of one of the middle riffles, and the upstream end of the reach. All measurements are taken mid-channel, at a height of 30 cm above the water. The densiometer is leveled with the sampler's head just visible in the crotch of the V (Figure 7). The number of cross-hairs covered by shade are counted and recorded in four directions, pivoting around the densiometer: upstream, left bank, downstream, and right bank. Counts of zero indicate all observation points were unshaded; counts of 17 indicate cover intersected all observation points on the grid. Shade includes vegetation, living or dead, or a man-made structure.

4.8.2 Landscape Change

Significant landscape disturbances within the reach and surrounding area since the last site visit are documented and photographed (e.g., landslides, fires, flooding, beaver activity, restoration planting, logging, new buildings, roads, culverts, or other infrastructure). Crew members will estimate the approximate area affected and age of the landscape change and record these changes on the field sheet.

4.8.3 Noxious Weeds

King County's Noxious Weed program has identified a list of priority species to track in the field; the list is updated annually. Field crews receive weed identification training from Noxious Weed Program staff at the beginning of each field season, and throughout the season, staff carry field guides to aid identification. At each site, staff scan the reach and immediate riparian zone for presence of noxious weeds. Staff photograph priority species, estimate density, and report findings to the Noxious Weed Program via the King County Connect mobile app installed on the crew's field phones.

5.0 LABORATORY ANALYSIS

A contract laboratory is used to process macroinvertebrate samples. This section describes the general taxonomic laboratory procedures. Additional details are described in the SOPs provided by the contract laboratory.

5.1 Sample Delivery

Preserved samples are periodically shipped to or picked up by the taxonomic laboratory during the sampling season. Prior to transport, samples are boxed and inventoried to verify that all samples are included on the COC form.

5.2 Taxonomic Lab Procedures

The target number of organisms identified and counted per sample is at least 500. To achieve this, the taxonomic lab uses standard sorting protocols (Plotnikoff and Wiseman 2001) including a Caton (1991) subsampling device. Technicians pour whole samples into the device, randomly order the subsampling grids, and based on that order, pick out all organisms in each grid until at least 500 organisms are counted. After the 500th individual is counted, the technician will continue to count all remaining organisms in the grid to minimize bias and ensure densities can be calculated (i.e., # organisms/#grids). Once the 500+ subsample is complete, technicians will inspect any remaining sample material and note if any large or rare taxa are present but had not been included in the 500+ subsample.

Once sorted, certified taxonomists use dissecting scopes to identify all organisms to their appropriate taxonomic level consistent with Puget Sound Stream Benthos Fine Taxonomic Resolution requirements (Section 5.3). Taxonomists record the identification, counts, life stages, and information about the condition of specimens for each picked 500+ subsample.

Organisms that cannot be identified to taxonomic targets because of immaturity, poor condition, or lack of complete current regionally applicable published keys are left at appropriate taxonomic levels that are coarser than those specified under the Fine Taxonomic requirements. To obtain accuracy in richness measures, these organisms are designated as “not unique” if other specimens from the same group can be taken to target levels. Organisms designated as “unique” are those that can be definitively distinguished from other organisms in the sample. Large/Rare organisms are identified, and these are recorded with a count of “1”.

Identified organisms are preserved in 80% ethanol in labeled vials, and archived for five years. Quality assurance procedures are carried out for each sample to assess sorting efficiency, identification, and data entry.

5.3 Taxonomic Resolution

Taxonomic resolution specifies the target level of taxonomic classification for specific taxa. In the Puget Sound region three levels of standard taxonomic effort (STE) have been used: fine, medium and coarse (Table 6). Prior to 2012, King County used the coarse STE Table 6). However, since 2012, all samples have been identified to fine STE. B-IBI scores calculated in the PSSB account for differences in STE and should be comparable.

Table 6. Standard taxonomic effort levels for benthic sample identification.

Taxa	Taxonomic Effort		
	Fine	Medium	Coarse
Oligochaeta (segmented worms)	Subfamily/Genus	Family	Subclass
Acari (mites)	Genus	Subclass	Subclass
Gastropoda (snails)	Genus	Genus	Family
Dytiscidae (predaceous diving beetles)	Genus	Genus (adults) Family (larvae)	Family
Simuliidae (black flies)	Genus	Genus (larvae) Family (pupae)	Family
Chironomidae (midges)	Genus/Sp/Sp grp	Subfamily/tribe	Family
Trichoptera (Pupae only; caddisflies)	Genus/Sp/Sp grp	Family	Order

5.4 Reporting Methods

Taxonomic data are uploaded directly into the PSSB by the contract taxonomic laboratory (Section 6.1). Once uploaded, data are immediately accessible by King County staff for download. The contract taxonomic laboratory delivers a technical summary of each year's sample taxonomy and quality control results directly to the project managers.

6.0 DATA MANAGEMENT

6.1 Puget Sound Stream Benthos Database

Taxonomic data are entered, stored, and analyzed through the PSSB web site (PSSB; http://www.pugetsoundstreambenthos.org). The PSSB was developed through collaboration by multiple agencies (City of Seattle, King, Pierce, and Snohomish counties) to create a database system that allows benthic macroinvertebrate data sharing and provides tools for data analysis. The PSSB stores macroinvertebrate data and allows users to calculate B-IBI scores in a standardized way. This allows for consistent comparisons among sites, programs, and jurisdictions over time.

6.1.1 Lab Reporting

Benthic macroinvertebrate data can be uploaded into the PSSB using one of three methods. First, taxa data can be entered directly through the website interface, featuring dropdown menus for taxa and spaces for counts. Second, data may be batch uploaded using a designated spreadsheet format. Finally, data can be uploaded by taxonomic labs, which directly enter data into batch upload templates.

6.2 Habitat Database

Habitat data was historically entered into and maintained in a Microsoft Access database within King County servers. That database stores habitat information collected from 2002-2016.

In 2017, King County's GIS Center created a new enterprise geodatabase "BIBIHabitat" to store habitat data. The geodatabase resides on GIS Center's GISSQLKC server. The data are entered using Esri's Web AppBuilder's Edit widget. The data can be viewed in ArcMap.

7.0 QUALITY CONTROL

7.1 Field Sampling

Field sampling quality control (QC) is accomplished and assessed through a variety of techniques that are described in the following sections.

7.1.1 Macroinvertebrate Sample Collection

To ensure a macroinvertebrate sample adequately represents communities present in riffles across the site, staff collect two samples from each of four riffles. If there are fewer than four riffles within the reach King County has permission to sample, staff should sample as many riffles as possible to ensure the sample is representative of communities within the accessible reach.

The net should be settled, flush with the substrate, as quickly as possible to avoid loss of individuals from the sample area. Likewise, to avoid collecting individuals from outside the sample area, staff should avoid disturbing the substrate upstream of the net and limit each 1-ft^2 sample collection to no more than 2 minutes (e.g., hand-scrubbing rocks for ≤1 minute, and agitation with garden tool = 1 minute).

Prior to discarding any large rocks collected in the net, staff should carefully wash them into the 500-µm mesh net or sieve to avoid losing macroinvertebrates. Similarly, staff should carefully inspect the net to be sure all macroinvertebrates inside the net are transferred to the sample bottle. Staff should also inspect all other equipment used to transfer the sample (e.g., sieve, tweezers, tub) to be sure all macroinvertebrates are placed in the sample bottle.

Samples are labeled with internal and external labels that include sample name, date, the collector's initials, and the total number of bottles per sample. This information is also recorded on a chain of custody form, and rechecked when the samples are boxed and sent to the taxonomic lab.

7.1.2 Habitat Data Collection and Entry

Staff communicate with each other at the site and refer to protocols, if needed, when determining if the reach area has suitable riffle habitat and is suitable for sample collection. When complete, the second crew member checks the habitat sample data sheet for accuracy and completeness before leaving the site.

To reduce the number of entry errors in the habitat database, a number of automated checks have been incorporated to alert staff to incomplete or invalid entries. These automated checks alerts occur nightly, and are sent to the staff who manage the database. In addition, each week during the sampling season, a subset of entered data (at least 10%)

will be evaluated against the raw data sheets. If errors are found in more than 10% of those checked, all entries will be checked.

7.2 Replicate Sampling

Replicate macroinvertebrate samples and replicate habitat data are collected at approximately 10% of the sites each year. These sites are referred to as "QC sites", and are selected randomly at the beginning of each field season.

Replicate macroinvertebrate samples are collected to evaluate intra-site variability. The replicate and primary samples are collected on the same day, and if possible, from the same riffles by two different crew members with separate Surber samplers. If riffles are wide enough, samples can be collected side-by-side, making sure not to influence each other's sampling efforts or disturb sediment upstream of each sample. If the stream is narrow or contains limited riffle habitat, samples may be collected one at a time, with the primary and replicate samples leapfrogging each other up the stream reach, being certain not to disturb the sediment or water upstream of the sampler or to overlap sample collection in the same area.

Replicate habitat data are also collected at the QC sites. Two crew members each independently collect habitat data. The data are subsequently compared to ensure collection methods are consistently applied across field staff.

7.3 Lab Analysis

QC procedures are performed by the contract laboratory to ensure accurate and precise picking, counting, taxonomic identification, and upload of data. QC procedures associated with initial picking and subsampling involves checking sorting efficiency. These checks are conducted on a random selection of 10% of the samples by independent observers who re-examine 100% of sorted substrate from each sample.

7.3.1 Subsampling

The QC technician re-examines all of the sorted substrate from 10% of the processed subsamples. The technician records and counts any organisms that were missed and adds these to the total number obtained in the original sort. Technicians calculate sorting efficiency (SE) as the total number of specimens in the first sort, divided by the total number of specimens in the second sort, and expressed as a percentage.

Quality control procedures for taxonomic determinations involves checking accuracy, precision, and enumeration. 10% of samples are randomly selected and all organisms re-identified and counted by an independent taxonomist. Taxa lists and enumerations are compared by calculating a Bray-Curtis similarity statistic (Bray and Curtis 1957), Percent Taxonomic Disagreement (PTD) and Percent Difference in Enumeration (PDE). Routinely, discrepancies between the original identifications and the QC identifications are discussed among the taxonomists, and necessary rectifications to the data are made. Discrepancies that cannot be rectified by discussions are routinely sent out to independent taxonomic specialists for identification.

7.3.2 Taxonomic Sample Data Entry

Taxonomic data are initially entered into the contract laboratory's database. Data entries are checked and then data are uploaded to the PSSB database. PSSB data entry error is limited by multiple QC features in the data entry tool. The data entry interface design includes standardized taxonomic names, station names, and sample IDs, thereby preventing misspellings. In addition, the data entry tool will not accept duplicate entries of project names, site ID's or taxon names. These two features prevent the most common data entry errors. The data entry screen also provides a total count for each sample which the data entry operator cross-checks against information provided on the original data sheet. If discrepancies are found, the error source is identified and corrected. Sometimes the source is due to summation errors in the source file. This ensures that the number of individuals entered for a sample was accurate. Quality assurance for taxonomic name accuracy is incorporated as a 5% review of raw taxonomic data after uploading.

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Appendix A. Changes in Methods and Sites from the 2002 SAP

Changes made to the monitoring program since 2002 are noted below.

Sites Dropped since 2002

Original ambient monitoring sites that have been dropped from the program.

Site Code	Latitude	Longitude	Stream	Last year sampled	Number of years sampled (2002-2020)	Reason site dropped
08BEA3325	47.759877	-122.09885	Daniels Creek	2007	3	Low flow
08BEA3478	47.694994	-122.08918	Mackey Creek	2015	13	No suitable riffle habitat
08CED4115	47.414139	-122.01711	Taylor Creek/Jem Creek (Lower Cedar)	2002	1	Low flow
08CED4479	47.384812	-121.98924	Walsh Lake Diversion	2009	7	Construction diverted flow permanently
08EAS1502	47.5578	-122.2297	Lake Washington tributary (Mercer Is.)	2011	5	Low flow
08EVA3474	47.678827	-122.09225	Evans Creek	2002	1	Site replaced by other site downstream
08EVA3897	47.651634	-122.05041	Evans Creek	2007	3	Property access issues and no suitable riffle habitat
08EVA4077	47.651647	-122.04153	Rutherford Creek	2014	7	Dry
08EVA4165	47.65009	-122.02643	Evans Creek tributary (0112)	2007	1	Dry
08EVA4249	47.63898	-122.02428	Evans Creek tributary (0111A)	2006	4	Property access issues
08ISS4373	47.501198	-122.02214	Issaquah Creek tributary (0203)	2011	6	Low flow
08LAK3616	47.553342	-122.0682	Tibbetts Creek	2016	11	Beaver dams eliminated riffle habitat
08LIT2488	47.7566	-122.16629	Little Bear Creek	2014	9	No suitable riffle habitat, too deep
08LIT2876	47.799615	-122.14312	Cuttrhoat Creek (0083)	2008	2	Road construction
08NOR0001	47.805712	-122.27003	Scriber Lake Creek	2003	2	Dry
08NOR1100	47.819401	-122.27328	Golde Creek	2003	2	No suitable riffle habitat
08NOR1756	47.877597	-122.22413	North Creek	2017	11	Development caused the stream to go dry
08NOR2028	47.834452	-122.20348	Silver Creek	2008	4	Insufficient riffle habitat
08SAM2946	47.689498	-122.13974	Sammamish River tributary (0102)	2010	8	No suitable riffle habitat
08SAM2951	47.711586	-122.13219	High School Creek	2007	4	No suitable riffle habitat
08WES1036	47.512926	-122.26748	Mapes Creek	2011	9	No suitable habitat due to restoration
09BLA0813	47.444251	-122.20346	Panther Creek (Black River) tributary	2011	6	Low flow
09COV1753	47.311991	-122.02539	Rock Creek (Covington)	2002	1	No suitable riffle habitat
09COV1798	47.340638	-122.01759	Covington Creek	2012	9	No suitable riffle habitat
09DEE2266	47.289178	-121.90794	Deep Creek (Green River)	2005	3	Denied access
09DUW0024	47.557851	-122.35329	Duwamish River tributary (Puget Park)	2007	2	No suitable riffle habitat, low flow
09DUW0144	47.514956	-122.30945	Hamm Creek	2011	8	No suitable riffle habitat
09JEN1358	47.373126	-122.09716	Jenkins Creek	2012	9	No suitable riffle habitat, too deep
09LOW0406	47.375221	-122.26581	Mullen Slough	2011	8	No suitable riffle habitat
09LOW0788	47.325585	-122.20249	Green River- Lower tributary (0069)	2014	11	No suitable riffle habitat
09MID1495	47.288025	-122.06737	Crisp Creek	2011	9	No suitable riffle habitat
09MID1704	47.273393	-122.02778	Green River - Middle tributary	2011	8	Low flow
09MIL0291	47.307684	-122.27607	Mill Creek (Auburn)	2003	2	No suitable riffle habitat
09MIL0497	47.365688	-122.2516	Mill Creek (Auburn)	2006	1	No suitable riffle habitat
09NEW1875	47.220818	-121.99785	Newaukum Creek	2013	9	High flow
09NEW1911	47.225585	-121.99065	Newaukum Creek	2009	6	High flow
09NEW2078	47.247236	-121.95461	Stonequarry Creek	2008	3	No suitable riffle habitat
09SOO1020	47.321254	-122.16028	Soosette Creek	2003	2	Property access issues
09SOO1040	47.417201	-122.15879	Big Soos Creek	2002	1	No suitable riffle habitat
09SOO1106	47.372486	-122.14803	Meridian Valley Creek	2011	7	Property access issues
09SOO1144	47.383312	-122.14054	Big Soos Creek	2011	7	No suitable riffle habitat

Changes Made to Habitat Survey Methods

Throughout the course of the program, habitat data collection and subsequent recording methods have been modified between most sampling seasons. These changes were made to facilitate more logical and precise data collection, recording, and entry. Aside from this, changes were also made to increase sampling efficiency, while reducing potential error and bias.

Changes in 2005

- Dominant riparian vegetation was changed to include a space for percent cover.
- Riparian vegetation classes were changed to include agriculture and herbaceous vegetation.
- Definition of shrubs was changed from being classified as woody stemmed vegetation that does not reach greater than 40 feet to woody stemmed vegetation less than 20 feet high.
- The size of the riparian buffer on each bank was classified as being 0', 1-10', 11-25', 25-50', and greater than 50 feet.
- Large woody debris (LWD) assessment was scaled back between 2003 and 2005. In 2003, rootwad width, rootwad stem, LWD length, circumference, diameter, contact with stream, and orientation were measured. Beginning in 2005, counts were taken of all LWD greater than six feet in length and four inches in diameter within the reach, as well as counts for root wads.
- Pebble counts were changed from sampling 35 points in each of the three riffles to sampling 100 points in the middle riffle. Pebble categories were expanded as well to differentiate the boulder category into small, medium, and large boulders.

Changes in 2006

- Additional fields were added to the site information to designate whether the site was a replicate and if the sampling location had changed from the previous year.
- Weather conditions were changed from an open note field to checkboxes, and a checkbox for recent heavy rain prior to sampling was added.
- Fields for overall reach length, number of pools, and stream stability were added.
- Cover percentages were changed from a blank field to categories (0-25%, 25-50%, 50-75%, 75-100%).
- Yes/No boxes were added to record if the left and/or right bank are composed predominately of native vegetation, as well as if the riparian buffer shades the stream.

Changes in 2007/2008

- The Yes/No field distinguishing whether the riparian buffers are composed predominately of native vegetation was condensed to represent both banks instead of each bank separately.

Changes in 2009

- Data entry field was added to the datasheet to track data entry progress.
- Checkboxes were added to clarify whether the site sampled was part of the ambient, CAO, or UPD program.
- Note was added to the field "Has there been heavy rain in the last 7 days?" to define heavy rain as greater than 0.5" in the past 24 hours prior to sampling.

- An inches/decimal-feet conversion chart was added so that all habitat measurements were recorded in decimal feet.
- Notes were added to clarify the 4 classes of stream bank stability.

Changes in 2010

- A checkbox was added to designate if a sample was not taken to help track cases where sites were visited but deemed inadequate to sample.
- A box was added to document the number of riffles that the sample was taken from as 1, 2, or 3. This was done because in many cases the three riffles needed for a 3 square foot sample were not available, requiring they be taken from one or two riffles instead.
- A box was added to document the presence of other organisms in the sample that are not included in the B-IBI calculations. This included checkboxes for crayfish, mussels, and other (e.g. fish or tailed frogs).

Changes in 2011

- Bankfull depth was changed from a blank field for the measurement to three categories (<1', 1-3', or >3').
- The dominant vegetation cover classes were removed so that just the dominant type of habitat was recorded rather than the type and amount.
- The urban cover category was broken into residential (low impervious) and commercial/industrial (high impervious).
- Yes/No box was added to document signs of recent (within 1-2 years) landscape change, with a note option to describe the change if present.
- Staff looked for and recorded the presence of invasive species, including non-native blackberry, knotweed, reed canary grass, ivy, holly, and yellow archangel.
- "EPA grant" was added as one of the project options for the site type being sampled.

Changes in 2012

- Starting in 2012, staff sampled 8, 1-ft^2 samples, instead of 3, 1-ft^2 samples. The "number of riffles sampled" field was changed from 3 to 4 to reflect the change from 3 square feet of sampling in 3 riffles to 8 square feet in 4 riffles (King County 2014b).
- The "distance to nearest road crossing" field was also changed from noting whether the nearest crossing was u/s or d/s and how far, to 3 check boxes noting whether a road crossing was present 50 feet upstream, 50 feet downstream, or none within 50 feet of the sample reach.
- A more comprehensive checklist was added to document noxious weeds observed at the sample site. This list includes 32 common noxious weeds, and whether they are regulated in King County.
- Starting in 2012, samples were identified to the Fine Taxonomic resolution standards (see Table 6). Before 2012, samples were identified to the Coarse Taxonomic resolution.

Changes in 2013

- The pebble count was changed from the previous method of sampling 100 pebbles from a transect extending along the entire bankfull width, to sampling 100 pebbles from the wetted width. The purpose of this change was to avoid a biased representation of the stream bed composition that is often created by sand bars or sediment deposited in the bankfull floodplain.
- An additional check for NZMS was added to the protocol to help us better assess the location and spread of invasive mudsnails.

Changes in 2016

- Air and water temperature at the time of collection were no longer recorded.

Changes in 2017

- Staff measured canopy cover using a densiometer. A concave densiometer was used at the beginning of the season, but after a side-by-side comparison, crew switched to the convex densiometer to more closely follow protocols established by the Washington State Department of Ecology's Watershed Health Monitoring Program.

Changes in 2018

- Staff record BIBI sampler and habitat sampling crew.
- Current and past weather were combined into one field, including if it had rained within the past 24 hours and if there had been heavy rain (defined as >1.3 cm (0.5 inches) within 24 hours) in the past 7 days.
- Bank stability changed to a scale of 0-10 rather than 1-4 and was recorded for Right and Left Banks throughout the reach.
- The number of rootwads is no longer counted separately.
- The Reach length was redefined to incorporate the area between the first and last Surber sample area and changed from feet to metric (meters).
- Rather than take measurements of each riffle, the wetted width, bankfull width and bankfull depth were taken at the Downstream, quarter, three-quarter and Upstream ends of the reach, all in metric (meters).
- The flow time was also reduced to only one of the middle riffles rather than each riffle, taken three times over one measured length (in meters) to approximate flow for the entire reach.

Appendix B: Monitoring Sites sampled in 2019

WRIA	Basin	Site Code	Latitude	Longitude	Stream
7	Lower Snoqualmie Basin	07CHR045515	47.74605	-121.89852	Cherry Creek
7	Lower Snoqualmie Basin	07CHR070059	47.75381	-121.8941	Margaret Creek
7	Lower Snoqualmie Basin	07CHR234411	47.7417	-121.93982	Cherry Creek
7	Lower Snoqualmie Basin	07GRN022903	47.610886	-121.864122	Griffin Creek
7	Lower Snoqualmie Basin	07GRN077319	47.615841	-121.900707	Griffin Creek
7	Lower Snoqualmie Basin	07HAR025863	47.68279	-121.92049	Snoqualmie River tributary (Harris Creek subbasin)
7	Lower Snoqualmie Basin	07HAR055371	47.726195	-121.879791	Harris Creek
7	Lower Snoqualmie Basin	07HAR070087	47.65292	-121.96087	Ames Creek
7	Snoqualmie Forks Basin	07LMS026663	47.544336	-121.543278	Snoqualmie River - Middle Fork tributary (Lower)
7	Snoqualmie Forks Basin	07LMS033235	47.51684	-121.61166	Snoqualmie River - Middle Fork tributary (Lower)
7	Snoqualmie Forks Basin	07LMS055075	47.488376	-121.719565	Snoqualmie River - Middle Fork tributary (Lower)
7	Snoqualmie Forks Basin	07LMS093027	47.48627	-121.757711	Snoqualmie River - Middle Fork tributary (Lower)
7	Snoqualmie Forks Basin	07LMS103591	47.54324	-121.54146	Snoqualmie River - Middle Fork tributary (Lower)
7	Snoqualmie Forks Basin	07LMS213875	47.495175	-121.640286	Snoqualmie River - Middle Fork tributary (Lower)
7	Lower North Fork Tolt River Basin-Tolt River	07LNT062039	47.722639	-121.739274	Tolt River
7	Lower North Fork Tolt River Basin-Tolt River	07LNT075179	47.74339	-121.708721	Tolt River
7	Lower North Fork Tolt River Basin-Tolt River	07LNT130827	47.744648	-121.705084	Tolt River
7	Lower Snoqualmie Basin	07LNT163403	47.744299	-121.820777	Lower S Fork Tolt River tributary (0303)
7	Lower South Fork Tolt Basin	07LST069191	47.688545	-121.786779	Tolt River
7	Lower Snoqualmie Basin	07PAT001415	47.56475	-121.853444	Mud Creek
7	Lower Snoqualmie Basin	07PAT166087	47.583426	-121.951276	Canyon Creek (Snoqualmie River)
7	Lower Snoqualmie Basin	07PAT187623	47.582202	-121.929938	Patterson Creek
7	Lower Snoqualmie Basin	07PAT228627	47.523697	-121.844634	Coal Creek (Snoqualmie River)
7	Lower Snoqualmie Basin	07PAT231079	47.54926	-121.88884	Snoqualmie River tributary (0430)
7	Lower Snoqualmie Basin	07RAG002867	47.498716	-121.91917	Snoqualmie River tributary (Raging River subbasin)
7	Raging River Basin	07RAG004615	47.547016	-121.90215	Raging River
7	Lower Snoqualmie Basin	07RAG018227	47.47915	-121.892999	Raging River tributary
7	Lower Snoqualmie Basin	07RAG022147	47.457368	-121.836318	Snoqualmie River tributary (Raging River subbasin)
7	Raging River Basin	07RAG042787	47.479864	-121.87046	Raging River
7	Raging River Basin	07RAG043443	47.497844	-121.905615	Raging River
7	Snoqualmie Forks Basin	07SFS009443	47.467888	-121.752865	Snoqualmie River - S Fork tributary
7	Snoqualmie Forks Basin	07SFS011059	47.488057	-121.785286	Snoqualmie River - S Fork tributary
7	Snoqualmie Forks Basin	07SFS011475	47.440617	-121.66643	Snoqualmie River - S Fork tributary

WRIA	Basin	Site Code	Latitude	Longitude	Stream
7	Snoqualmie Forks Basin	07SFS041891	47.470956	-121.772703	Snoqualmie River - S Fork tributary
7	Snoqualmie Forks Basin	07SFS066611	47.440732	-121.649546	Snoqualmie River - S Fork tributary
7	Stossel Creek - Tolt River Basin	07STL080295	47.63797	-121.914658	Tolt River
7	Stossel Creek - Tolt River Basin	07STL184007	47.66959	-121.86469	Tolt River
7	Lower Snoqualmie Basin	07TKL047047	47.5802	-121.78183	Tokul Creek
7	Lower Snoqualmie Basin	07TKL072439	47.621687	-121.784871	Tokul Creek
8	Sammamish River Basin	08BEA3312	47.703496	-122.09254	Monticello Creek
8	Sammamish River Basin	08BEA3321	47.754916	-122.104024	Cold Creek
8	Sammamish River Basin	08BEA3474	47.677597	-122.098301	Bear Creek (Sammamish River)
8	Sammamish River Basin	08BEA3571	47.744769	-122.088883	Cottage Lake Creek
8	Sammamish River Basin	08BEA3650	47.717871	-122.076997	Bear Creek (Sammamish River)
8	Sammamish River Basin	08BEA3737	47.718328	-122.073941	Seidel Creek
8	Sammamish River Basin	08BEA3747	47.758758	-122.057993	Bear Creek (Sammamish River)
8	Sammamish River Basin	08BEA3826	47.7336	-122.05881	Struve Creek
8	Sammamish River Basin	08BEA3914	47.73302	-122.05998	Colin Creek
8	Cedar River / Lake Washington Basin	08CED2433	47.474227	-122.163593	Maplewood Creek
8	Cedar River / Lake Washington Basin	08CED2518	47.466198	-122.159083	Molasses Creek
8	Cedar River / Lake Washington Basin	08CED2711	47.453235	-122.137979	Madsen Creek
8	Cedar River / Lake Washington Basin	08CED2898	47.461466	-122.125545	Cedar River - Lower tributary (0311)
8	Cedar River / Lake Washington Basin	08CED4192	47.374751	-122.017672	Rock Creek (Lower Cedar)
8	Cedar River / Lake Washington Basin	08CED4975	47.410569	-121.921792	Hotel Creek (0342)
8	Cedar River / Lake Washington Basin	08CED5032	47.415086	-121.887131	Rock Creek (Upper Cedar)
8	Cedar River / Lake Washington Basin	08CED5046	47.4277	-121.91545	Webster Creek - Upper (0341)
8	Cedar River / Lake Washington Basin	08EAS1536	47.709929	-122.248781	Denny Creek
8	Cedar River / Lake Washington Basin	08EAS1964	47.529663	-122.200605	May Creek (Lake Washington)
8	Cedar River / Lake Washington Basin	08EAS2058	47.519171	-122.194691	May Creek (Lake Washington)
8	Cedar River / Lake Washington Basin	08EAS2191	47.695551	-122.197014	Forbes Creek (0242)
8	Cedar River / Lake Washington Basin	08EAS2272	47.623286	-122.156843	Kelsey Creek
8	Cedar River / Lake Washington Basin	08EAS2446	47.55253	-122.16512	Coal Creek (Lake Wash.)
8	Cedar River / Lake Washington Basin	08EAS2540	47.550172	-122.156867	Coal Creek (Lake Wash.) tributary (0273)
8	Cedar River / Lake Washington Basin	08EAS2546	47.582723	-122.157426	Sunset Creek
8	Cedar River / Lake Washington Basin	08EAS2631	47.518948	-122.163144	May Creek (Lake Washington)
8	Sammamish River Basin	08EVA3555	47.67462	-122.081052	Evans Creek
8	Sammamish River Basin	08EVA3637	47.65296	-122.06841	Evans Creek tributary (0111E)
8	Sammamish River Basin	08EVA3640	47.674662	-122.072706	Evans Creek tributary (0108)
8	Sammamish River Basin	08EVA3642	47.67972	-122.07785	Evans Creek tributary (0107)
8	Sammamish River Basin	08EVA3813	47.67341	-122.063447	Evans Creek tributary (0108A)

WRIA	Basin	Site Code	Latitude	Longitude	Stream
8	Sammamish River Basin	08ISS3877	47.551207	-122.046876	Issaquah Creek
8	Sammamish River Basin	08ISS3958	47.518325	-122.036396	Issaquah Creek tributary (0198)
8	Sammamish River Basin	08ISS3962	47.538939	-122.0426	Issaquah Creek
8	Sammamish River Basin	08ISS4294	47.484906	-122.028632	Fifteenmile Creek
8	Sammamish River Basin	08ISS4573	47.531946	-121.976205	High Point Creek
8	Sammamish River Basin	08ISS4724	47.427716	-121.978422	Carey Creek
8	Sammamish River Basin	08ISS4730	47.433941	-121.971913	Holder Creek
8	Sammamish River Basin	08ISS4735	47.47075	-121.964356	Holder Creek tributary
8	Sammamish River Basin	08ISS4748	47.531728	-121.983455	Issaquah Creek - E Fork
8	Sammamish River Basin	08ISS4884	47.525851	-121.941132	Issaquah Creek - E Fork
8	Sammamish River Basin	08LAK2827	47.578488	-122.113038	Vasa Creek
8	Sammamish River Basin	08LAK3121	47.641052	-122.106516	Idylwood Creek
8	Sammamish River Basin	08LAK3540	47.600939	-122.080335	Pine Lake Creek
8	Sammamish River Basin	08LAK3627	47.608584	-122.071866	Ebright Creek
8	Sammamish River Basin	08LAK3628	47.615284	-122.066971	Eden/George Davis Creek
8	Sammamish River Basin	08LAK3699	47.541782	-122.064195	Tibbetts Creek - Lower
8	Sammamish River Basin	08LAK3879	47.565113	-122.050183	Laughing Jacobs Creek
8	Sammamish River Basin	08LAK3880	47.569898	-122.053288	Many Springs Creek
8	Sammamish River Basin	08LIT2585	47.758429	-122.160347	Little Bear Creek
8	Sammamish River Basin	08LIT2602	47.83994	-122.16225	Little Bear Creek
8	Sammamish River Basin	08LIT2603	47.847646	-122.163836	Little Bear Creek
8	Sammamish River Basin	08LIT2682	47.763211	-122.156929	Little Bear Creek
8	Sammamish River Basin	08LIT2685	47.778471	-122.154549	Little Bear Creek
8	Sammamish River Basin	08LIT2692	47.811464	-122.158997	Little Bear Creek
8	Sammamish River Basin	08LIT2781	47.790039	-122.144532	Little Bear Creek
8	Sammamish River Basin	08NOR1362	47.779822	-122.250431	Swamp Creek
8	Sammamish River Basin	08NOR1370	47.770053	-122.244606	Swamp Creek
8	Sammamish River Basin	08NOR1750	47.852841	-122.222496	North Creek
8	Sammamish River Basin	08NOR2115	47.791242	-122.195243	North Creek tributary
8	Sammamish River Basin	08NOR2306	47.778784	-122.187628	North Creek
8	Sammamish River Basin	08SAM0000	47.682628	-122.143714	Peters Creek
8	Sammamish River Basin	08SAM1914	47.745483	-122.211857	Sammamish River tributary (0068)
8	Sammamish River Basin	08SAM2674	47.72908	-122.15256	Sammamish River tributary (0096D)
8	Sammamish River Basin	08SAM2855	47.696997	-122.131529	Sammamish River tributary (0101)
8	Sammamish River Basin	08SAM2862	47.733381	-122.13722	Sammamish River tributary (0090)
8	Sammamish River Basin	08SAM2865	47.742702	-122.141764	Gold Creek
8	Sammamish River Basin	08SAM3045	47.712241	-122.12057	Sammamish River tributary (0095F)

WRIA	Basin	Site Code	Latitude	Longitude	Stream
8	Sammamish River Basin	08SAM3047	47.720449	-122.124316	Sammamish River tributary (0095D)
8	Cedar River / Lake Washington Basin	08WES0622	47.673452	-122.310727	Ravenna Creek
8	Cedar River / Lake Washington Basin	08WES0629	47.702708	-122.308342	Maple Leaf Creek
8	Cedar River / Lake Washington Basin	08WES0903	47.752199	-122.281877	McAleer Creek
8	Cedar River / Lake Washington Basin	08WES0905	47.759565	-122.28683	Lyon Creek
8	Cedar River / Lake Washington Basin	08WES1037	47.518425	-122.268797	Mapes Creek
8	Cedar River / Lake Washington Basin	08WES1178	47.766499	-122.263888	Lake Washington tributary (0056)
8	Cedar River / Lake Washington Basin	08WES1340	47.507869	-122.247582	Taylor Creek (W Lake Washington)
8	Cedar River / Lake Washington Basin	08WES1490	47.503982	-122.227702	Lake Washington tributary (0464B)
8	Cedar River / Lake Washington Basin	08WES1579	47.483496	-122.227268	Lake Washington tributary
9	Duwamish - Green River Basin	09BLA0675	47.381499	-122.221081	Mill Creek (Kent)
9	Duwamish - Green River Basin	09BLA0716	47.382722	-122.223995	Mill Creek (Kent)
9	Duwamish - Green River Basin	09BLA0722	47.407005	-122.216196	Garrison Creek - S Fork
9	Duwamish - Green River Basin	09BLA0756	47.369338	-122.208823	Mill Creek (Kent)
9	Duwamish - Green River Basin	09BLA0768	47.428348	-122.214069	Springbrook Creek (Black River)
9	Duwamish - Green River Basin	09BLA0771	47.442177	-122.207574	Panther Creek (Black River)
9	Duwamish - Green River Basin	09BLA0772	47.445022	-122.211052	Panther Creek (Black River)
9	Duwamish - Green River Basin	09BLA0817	47.46604	-122.207183	Springbrook Creek tributary (0006B)
9	Duwamish - Green River Basin	09COV1165	47.31919	-122.11905	Covington Creek
9	Duwamish - Green River Basin	09COV1418	47.30933	-122.077692	Covington Creek
9	Duwamish - Green River Basin	09COV1756	47.32877	-122.022072	Covington Creek
9	Duwamish - Green River Basin	09COV1862	47.317211	-122.00522	Rock Creek tributary (Covington)
9	Duwamish - Green River Basin	09COV1864	47.325762	-122.001348	Rock Creek tributary (Covington)
9	Duwamish - Green River Basin	09DEE2163	47.282302	-121.932687	Deep Creek (Green River)
9	Duwamish - Green River Basin	09DEE2208	47.266907	-121.916937	Coal Creek (Green River)
9	Duwamish - Green River Basin	09DEE2211	47.285142	-121.923905	Deep Creek (Green River)
9	Duwamish - Green River Basin	09DEE2294	47.275939	-121.894775	Coal Creek (Green River)
9	Duwamish - Green River Basin	09DUW0091	47.510357	-122.322142	Duwamish River tributary (0001E)
9	Duwamish - Green River Basin	09DUW0225	47.502212	-122.300156	Duwamish River tributary (0003)
9	Duwamish - Green River Basin	09DUW0277	47.489932	-122.289451	Riverton Creek (003D)
9	Duwamish - Green River Basin	09JEN1318	47.362218	-122.099903	Jenkins Creek
9	Duwamish - Green River Basin	09JEN1357	47.3689	-122.098899	Jenkins Creek
9	Duwamish - Green River Basin	09LOW0325	47.464905	-122.281488	Lower Green River tributary (0036B)
9	Duwamish - Green River Basin	09LOW0751	47.343487	-122.20504	Olson Creek
9	Duwamish - Green River Basin	09LOW0753	47.353074	-122.20751	Green River - Lower tributary (S 277th St.)
9	Duwamish - Green River Basin	09MID1374	47.275597	-122.088114	O'Grady Creek
9	Duwamish - Green River Basin	09MID1537	47.291898	-122.059923	Crisp Creek tributary

WRIA	Basin	Site Code	Latitude	Longitude	Stream
9	Duwamish - Green River Basin	09MID1744	47.272614	-122.021072	Cristy Creek
9	Duwamish - Green River Basin	09MID1817	47.26799	-122.009736	Cristy Creek
9	Duwamish - Green River Basin	09MID1958	47.278886	-121.978571	Icy Creek
9	Duwamish - Green River Basin	09MID2426	47.315006	-121.867614	Green River - Middle tributary
9	Duwamish - Green River Basin	09MIL0340	47.303209	-122.272324	Mill Creek (Auburn)
9	Duwamish - Green River Basin	09MIL0390	47.303166	-122.260837	Mill Creek (Auburn)
9	Duwamish - Green River Basin	09NEW1657	47.250042	-122.037744	Newaukum Creek
9	Duwamish - Green River Basin	09NEW2076	47.235936	-121.950976	Newaukum Creek - N Fork
9	Duwamish - Green River Basin	09NEW2102	47.231078	-121.94603	Newaukum Creek
9	Duwamish - Green River Basin	09NEW2128	47.234245	-121.93519	Newaukum Creek - N Fork
9	Duwamish - Green River Basin	09NEW2151	47.224415	-121.931257	Newaukum Creek
9	Duwamish - Green River Basin	09SOO0943	47.30855	-122.16904	Big Soos Creek
9	Duwamish - Green River Basin	09SOO1022	47.331725	-122.155276	Soosette Creek
9	Duwamish - Green River Basin	09SOO1130	47.31817	-122.135959	Soos Creek
9	Duwamish - Green River Basin	09SOO1134	47.336409	-122.135101	Soos Creek
9	Duwamish - Green River Basin	09SOO1209	47.358599	-122.125122	Little Soos Creek
9	Duwamish - Green River Basin	09SOO1283	47.372825	-122.113902	Little Soos Creek
10	Puyallup-White	BSE_1_MudMtnRd	47.175524	-122.018775	Boise Creek
10	Puyallup-White	BSE_21_GolfCrs	47.195154	-121.953301	Boise Creek
10	Puyallup-White	BSE_8_268thAveSE	47.190111	-121.981778	Boise Creek
9/15	Duwamish - Green River Basin/Vashon	VashChris	47.40277	-122.516930	Christenson Creek
9/15	Duwamish - Green River Basin/Vashon	VashShing	47.47628	-122.481500	Shinglemill Creek
9/15	Duwamish - Green River Basin/Vashon	28A	47.403459	-122.468866	Judd Creek

Appendix C: Habitat Data Sheet

The following habitat sheet was used for the 2019 field season. Modifications are made as needed, including an annual update of the noxious weeds section.

☐ **Did NOT Sample** ☐ **Ambient** ☐ **Miller/Walker** ☐ **Vashon** ☐ **Boise** ☐ **Stormwater Retrofit** ☐ **Other:** ___________

Site Name/Number:	Replicate site? **Yes** **No**	**Data Entry-** *check when complete*
Date: Time:	BIBI Sampler: Habitat Sampler:	☐ **Habitat** *Date:*_____ *Initials:*_____ | ☐ **PSSB** *Date:*_____ *Initials:*_____ | ☐ **Photos** *Date:*_____ *Initials:*_____ | ☐ **Site-Master** *Date:*_____ *Initials:*_____
Location:	Changed location? **Y** **N** *If yes, explain:*	

NEAREST ROAD X-ING
Within 15 m of reach:
☐ Upstream
☐ Downstream
☐ None within 15 m

WEATHER Currently:
☐ Sunny
☐ Partly cloudy (10 – 50%)
☐ Mostly cloudy (>50%)
☐ Raining

Past:
☐ Rained in the last 24 hours?
☐ Heavy rain in last 7 days? (>1.3 cm (0.5") in 24 hours)
☐ N/A

WATER CONDITIONS
☐ Clear
☐ Turbid

BANK STABILITY *(circle one for each bank, facing DS)*	**Armoring Present?**	**Bank stable** Evidence of erosion or bank failure absent or minimal; little potential for future problems. <5% of bank affected.	**Moderately stable** Infrequent, small areas of erosion mostly healed over. 5-30% of bank in reach has areas of erosion.	**Moderately unstable** 30-60% of bank in reach has areas of erosion; high erosion potential during floods.	**Unstable** Many eroded areas; "raw" areas frequent along straight sections and bends; obvious bank sloughing; 60-100% of bank has erosional scars.
Right Bank	**Y N**	**10 9**	**8 7 6**	**5 4 3**	**2 1 0**
Left Bank	**Y N**	**10 9**	**8 7 6**	**5 4 3**	**2 1 0**

REACH LENGTH
Top of u/s riffle to bottom of d/s riffle
__________ m

SAMPLED FROM *(circle one)*
1 2 3 4 riffles

LARGE WOODY DEBRIS
(>2 m. long & >10 cm. dia., OR rootwads >0.3 m^3)
\# within BFW: ___________ ☐ no LWD in reach

 # POOLS: ________

WETTED WIDTH ___________m	WETTED WIDTH ___________m	WETTED WIDTH ___________m	WETTED WIDTH ___________m
BANKFULL WIDTH ___________m	BANKFULL WIDTH ___________m	BANKFULL WIDTH ___________m	BANKFULL WIDTH ___________m
BANKFULL DEPTH ___________cm	BANKFULL DEPTH ___________cm	BANKFULL DEPTH ___________cm	BANKFULL DEPTH ___________cm
THALWEG DEPTH ___________cm	THALWEG DEPTH ___________cm	THALWEG DEPTH ___________cm	THALWEG DEPTH ___________cm

FLOW TIME
A middle riffle
Length:
_________**m**
Time:
_________**sec**
_________**sec**
_________**sec**

RIFFLE DEPTH
Average for each surber sample
__________**cm**
__________**cm**
__________**cm**
__________**cm**
__________**cm**
__________**cm**
__________**cm**
__________**cm**

OTHER CRITTERS * *Indicate # alive (a) or dead (d), photograph dead*
VERTEBRATES (in sample, remove) ☐ Salmonids* ___________
☐ Tailed Frog _____ ☐ Lamprey _____ ☐ Sculpin*__________
☐ Other______________ ☐ None
INVERTEBRATES (in sample)
☐ Mussels* (*DO NOT include* in sample) _______________
☐ Crayfish (*INCLUDE* in sample) ______ ☐ None

NZMS Check	Present? (Y/N)	Collected? (Y/N)	Habitat
Habitat #1			
Habitat #2			

Habitat Type: (Rf) – Riffle, (Rn) – Run, (P) – Pool
Infestation Levels (circle one): Trace Low Moderate High

NOTES:

Site:	Date:	Personnel:

Signs of recent landscape change (within 1-2 years)? *(Landslide, beaver, veg clearing, reveg, construction, etc. Yes: describe in notes, take photos)* Y N	*Priority Noxious Weeds Observed at Site*

☐ No listed species found

TOP PRIORITY
☐ Common reed
☐ European coltsfoot
☐ Garden loosestrife
☐ Garlic mustard
☐ Giant hogweed
☐ Knapweed +
☐ Poison-hemlock
☐ Purple loosestrife
☐ Policeman's helmet +
☐ Tansy ragwort +

☐ Submitted to Report A Weed or King County Connect?

+ Pull and bag plants if population is small

OTHER species of concern (report on public properties only)
☐ Butterfly bush
☐ English ivy
☐ English holly
☐ Italian arum
☐ Knotweed
☐ Multiflora rose
☐ Old man's beard
☐ Scotch broom
☐ Spotted jewelweed
☐ Yellow archangel
☐ Yellow flag iris
☐ Unknown (description, photos)
NOTES: Include estimated size/density, actions taken, **Take pictures!**

Photos?
(Site name, US, DS, RB, LB, USpano, DSpano, weeds, why not sampled, mudsnails, critters, landscape changes, garbage)
Y N

DENSIOMETER – DS and US ends of reach and middle of one of the middle riffles, representative of where samples were taken, 30 cm above water, center of riffle/channel. Record #covered cross-sections out of 17 for each direction (does not need to be cardinal directions).

DS		M		US	
US	_____	US	_____	US	_____
RB	_____	RB	_____	RB	_____
DS	_____	DS	_____	DS	_____
LB	_____	LB	_____	LB	_____

WOLMAN PEBBLE COUNT:

Collect from wetted width

Count at least 100 total.

Complete the full transect even after 100 is reached.

Middle Riffle unless otherwise noted.

Substrate Class	Size Class (mm)		Pebble Count	Total
Sand	0 - 2	A		
Very Fine Gravel	>2 - 2.8	B		
Very Fine Gravel	>2.8 - 4	C		
Fine Gravel	>4 - 5.6	D		
Fine Gravel	>5.6 - 8	E		
Medium Gravel	>8 - 11	F		
Medium Gravel	>11 - 16	G		
Coarse Gravel	>16 - 22.6	H		
Coarse Gravel	>22.6 - 32	I		
Very Coarse Gravel	>32 - 45	J		
Very Coarse Gravel	>45 - 64	K		
Small Cobble	>64 - 90	L		
Medium Cobble	>90 - 128	M		
Large Cobble	>128 - 180	N		
Very Large Cobble	>180 - 256	O		
Small Boulder	>256 - 512	P		
Medium Boulder	>512 - 1024	Q		
Very Large Boulder	>1024 - 2048	R		
			Grand Total (>100)	

Appendix D. Chain of Custody Form

The Chain of Custody (COC) form that will be used for the 2020 sampling season.

King County

2020 CHAIN OF CUSTODY RECORD

Benthic Macroinvertebrate Ambient Monitoring

Project Name: ______________________________

DNRP- Water and Land Resources Division
201 South Jackson Street - MS KSC NR 0600
Seattle, WA 98119

Contacts: **Kate Macneale** 206-477-4769

Sample ID	Collect Date mm/dd/yy	No. of Containers	QC rep?	Box No.	Collected by:
	/ / 20				
	/ / 20				
	/ / 20				
	/ / 20				
	/ / 20				
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	/ / 20				
Totals:					

Projects: Ambient (WRIA 8 & 9); WRIA 7 & Boise, Vashon, Seattle, Stormwater Retrofit, Miller/Walker, Other.
Ideally 1 project per chain of custody page

Staff initials/names: KHM - Kate Macneale, **BAS** - Beth Sosik, **HRF** - Houston Flores, **SAH** - Stephanie Hess, **NAC** - Natalie Carrigan

Specify others: