Integrated Ambient Monitoring Follow-up Study in Indian Creek

An Investigation of the Causes of Biological Impairment and a Further Demonstration of the Instream Monitoring Approach

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Cover photo: Upstream Indian Creek In-Situ Trout "Redd" (photo by Patti Sandvik)

Integrated Ambient Monitoring Follow-up Study in Indian Creek

Investigation of the Causes of Biological Impairment and Further Demonstration of the Instream Monitoring Approach

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Abstract

This 2013 study of Indian Creek, located in Olympia, Washington, extends the work conducted in 2010 (Marshall and Era-Miller, 2012). The 2010 study results indicated that lower Indian Creek would not support salmon reproduction. The lower site is in the midst of commercial buildings and parking lots. In 2013, the stormwater pipe suspected of discharging the pollutants causing mortalities to trout early lifestages was bracketed with additional monitoring stations.

The 2013 monitoring began with instream exposures of rainbow trout eyed-embryos in simulated redds and ended when the trout became swim-up fry. Survival just downstream of the stormwater pipe was 4% at the alevin lifestage. Survival of alevins just above the pipe was 60%. Fry survival 13 days later at the upper Indian Creek site was 93%. The upstream site has a wooded riparian buffer, along with nearby residential and commercial land uses and a highway (I-5).

Surviving trout were analyzed for six metals. Copper in fish tissue strongly correlated ($r = ^{-}0.99$) with fry survival. Both tissue zinc ($r = ^{-}0.87$) and copper ($r = ^{-}0.71$) correlated moderately with alevin survival. Evidence from tissue metals and stream PAHs indicates that metal and PAH mixtures contributed to trout mortality.

Periphyton and macroinvertebrates were assessed because they are the food chain base and provide sustenance for growing young salmon. Periphyton and macroinvertebrates were analyzed for metals to evaluate their usefulness for pollutant monitoring. Lower site benthic communities showed impairment, including an increase in metals-tolerant organisms.

Stream, stormwater, groundwater, and sediment samples were analyzed for metals and PAHs. Stream, stormwater, and groundwater samples were also analyzed for oxygenated (ketone- and quinone-substituted) PAHs. In addition, groundwater and sediment samples were analyzed for base/neutral acid extractable organics.

Results show lower Indian Creek to be unsuitable for salmon reproduction and the weight of evidence implicates a mixture of pollutants in the creek.

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Introduction

Study Concept

Focus of Study

Successful salmon reproduction is the most highly valued feature of a healthy stream in the Pacific Northwest. Adult salmon return from the ocean to spawn in urban rivers and streams, and their offspring must survive and develop within these urban areas until ocean migration. Protecting salmon early lifestages and the food on which they depend is the key to maintaining productive streams. Pacific Northwest fish populations are particularly susceptible to the toxic effects of urban stormwater runoff (Feist et al., 2011; Scholz et al., 2011).

McCarthy et al. (2008) considered the results of recent field work on coho prespawn mortalities, along with relevant contaminant-specific toxicological findings in seeking to understand the effect of stormwater on fish health in California and the Pacific Northwest. One of their conclusions was that exposure to complex mixtures from nonpoint sources was almost always the reality in streams and that the effects of these complex mixtures were hard to predict. Another conclusion was that biological monitoring must be a key component of stream restoration. The inability to predict toxicity based on the results of chemical analysis leads to the second conclusion about the necessity of biological monitoring in assessing water quality.

Salmon reproduction is the focus of the integrated ambient monitoring used in Indian Creek in 2010 and 2013. This focus also includes the stream primary producers (periphyton) and primary consumers (macroinvertebrates). Benthic macroinvertebrates feed on periphyton or detritus and are a key food source for fish in streams. The breadth of the focus means that monitoring results will be generally applicable and benefit other fish species as well. The approach remains affordable and manageable by not including every species directly. The use of the instream biological monitoring approach provides both environmental realism and efficiency.

Design of Current Study

The goal of the monitoring approach evaluated here is to assess the suitability of a stream to support salmon reproduction and thereby show whether pollution controls are adequate. The diagnostic ability of the monitoring approach is intended to show a path forward for further pollution controls, rather than reach a definite conclusion about the causes of instream toxicity. The 2013 study followed-up on the findings of a 2010 study which demonstrated that lower Indian Creek, an urban stream located in Olympia, Washington, is not a suitable stream for supporting salmon early lifestages or the macroinvertebrates they need for survival and growth (Marshall and Era-Miller, 2012). The 2013 study repeated the techniques found in the 2010 study to be the most informative and convenient.

We used rainbow trout (*Oncorhynchus mykiss*) eggs and alevins to test the in-situ toxicity of Indian Creek. Rainbow trout are in the Pacific salmon genus, *Oncorhynchus*, and are a suitable surrogate for other members of the genus. Rainbow trout eggs are readily available for use in an

early lifestage instream toxicity test. Test organisms placed in a stream experience a realistic environmental exposure and will respond to a broad spectrum of toxic chemicals and mixtures.

Biological assessments (bioassessments) directly measure the composition of communities of living organisms (macroinvertebrates, fish, or plants) to look for signs of water quality impairment. Benthic macroinvertebrates and periphyton are nearly stationary and experience a pollutant exposure that is representative of a particular location. This makes assessing these communities a useful tool for characterizing stream health at various locations.

Both the test trout exposed in the stream and the naturally occurring stream organisms will accumulate chemicals that can be measured in tissue samples and give an indication of pollutant exposure in the stream.

Ultimately, the routine application of this approach would be most useful when stormwater controls are nearing completion or before a stream in a developing area becomes polluted.

Methods

Study Area Description

The project focused on Indian Creek, an urban stream in the city of Olympia. Indian Creek is located in the South Puget Sound area and drains into Budd Inlet (Figure 1). The creek is 3 miles long, and its watershed is approximately 1,500 acres containing 35% impervious surface (Reynolds and Wood, 2011).

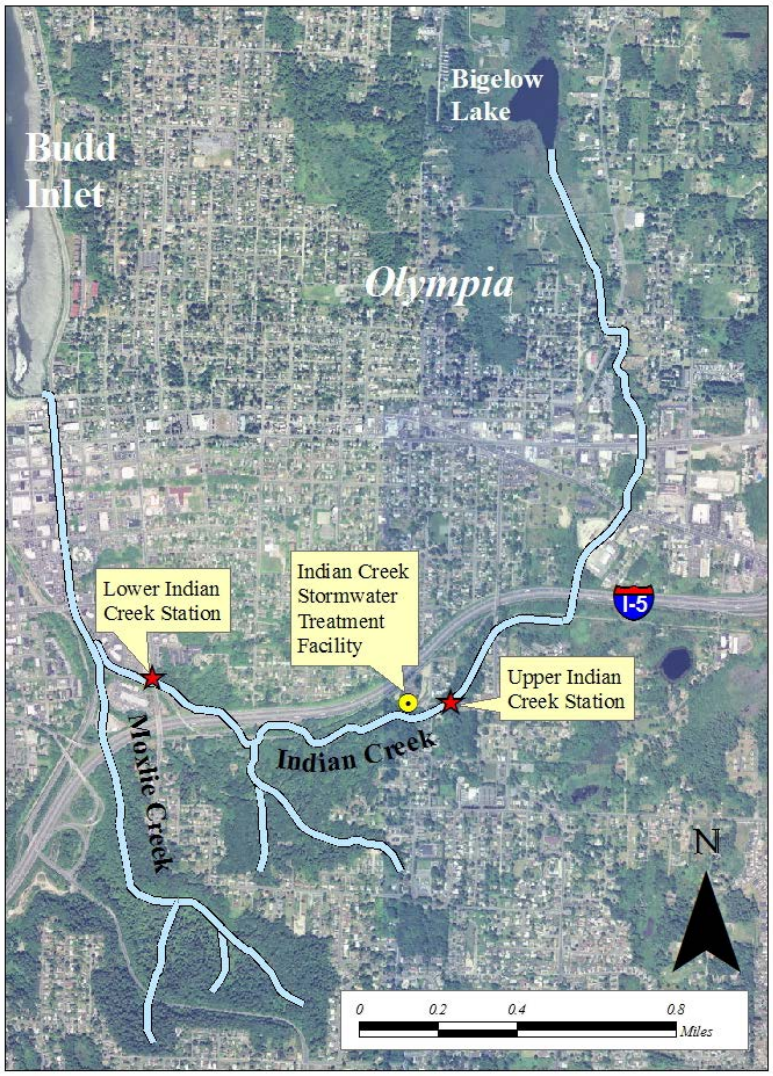

Figure 1. Indian Creek watershed and study locations.

Indian Creek originates from a wetland complex that includes Bigelow Lake and then flows through a mix of land uses including urban, industrial, residential, and parks. The creek crosses under Interstate 5 twice and under numerous other roads. It eventually joins Moxlie Creek and is then piped under downtown Olympia to the East Bay of Budd Inlet. Many of the culverts on Indian Creek are too small or have too much drop for salmon migration. Despite these barriers, resident trout inhabit the stream (City of Olympia, 2010). Numerous pollution sources, including the Indian Creek Stormwater Treatment Facility, drain into Indian Creek below the upper site.

The study's upstream site is in a wooded area, and the downstream site is in the midst of buildings and parking lots. Both are close to a busy interstate highway (I-5) (Figure 2 and 3). In response to the 2010 study results, additional monitoring stations were added to lower Indian Creek (Figure 4) in 2013 to refine understanding of the locations of pollutant sources.

Figure 2. Upper Indian Creek.

Figure 3. Lower Indian Creek.

Locations of Field Activities

The upper Indian Creek station is identified as I-1 in this study. It was called Indian 1 in the 2010 study report (Marshall and Era-Miller, 2012). The single lower Indian Creek station monitored in 2010 was called Indian 2. Two other stations were added to lower Indian Creek in 2013 to bracket the stormwater culvert suspected of being a significant source of toxicity in 2010. Station I-2A was added just upstream of the culvert, and I-2B was located just downstream of the culvert. Station I-2C was used in 2013 as a repeat of the location called Indian 2 in 2010. I-2B is closer to the stormwater pipe than the 2010 lower station now called I-2C. See Figure 4 and Table 1 for a diagram of the locations of the project activities in lower Indian Creek.

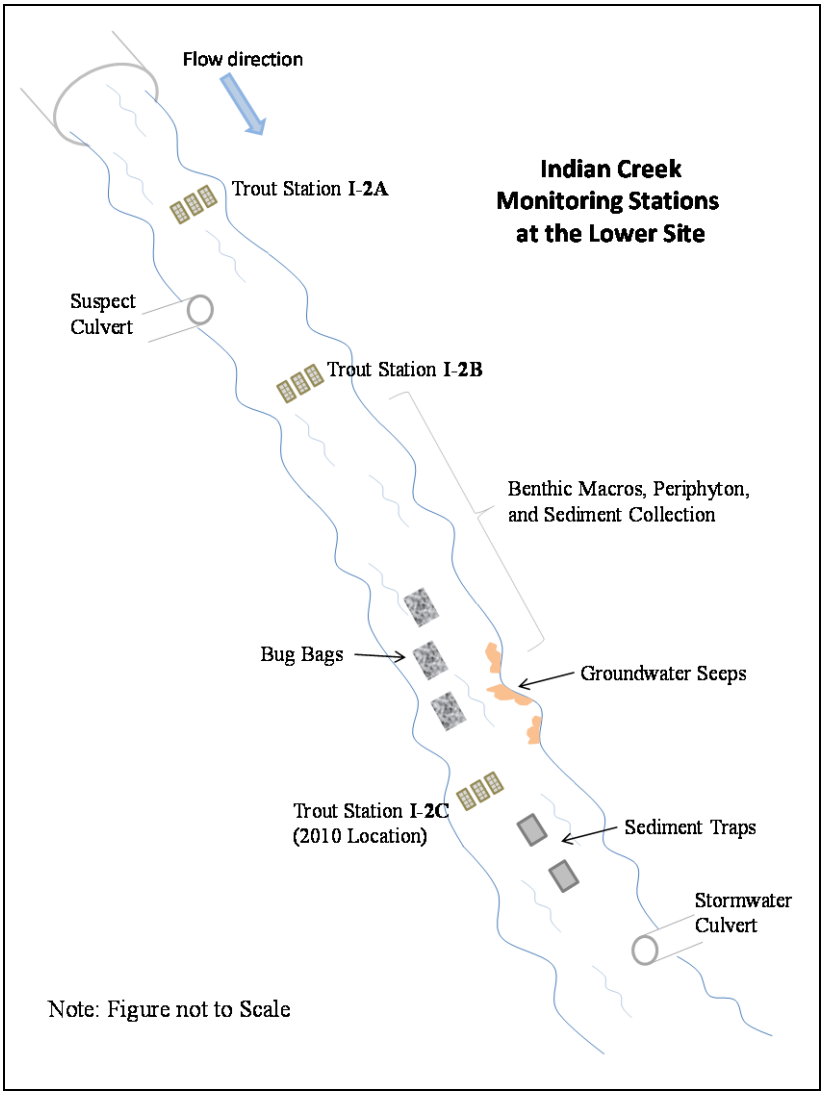

Figure 4. Detail of monitoring locations at lower Indian Creek.

Table 1. Relative location of lower Indian Creek field activities.

Below Eastside St. culvert		Below the next upstream site		Activity site
feet	meters	feet	meters	
33	10.1	33	10.1	I2-A
92	28.0	59	18.0	suspect stormwater culvert
128	39.0	36	11.0	I2-B
212	64.6	84	25.6	bug bags & groundwater seeps
226	68.9	14	4.3	I2-C
240	73.2	14	4.3	upper sediment trap
261	79.6	21	6.4	lower sediment trap
597	182.0	336	102.4	2nd stormwater culvert
697	212.4	100	30.5	back underground

Timing of Environmental Samples

The project took place during late spring of 2013 so the results would be comparable to the results of the 2010 study, which was also conducted in the late spring. Table 2 shows the timing of environmental sample collection.

Table 2. Timing of environmental sample collection.

Sample source	Date	Time	Location	Weather
surface water	4/23/2013	10:15	2010 passive sampler station	dry; rain the day before
	4/23/2013	10:55	duplicate	
	6/12/2013	15:22	2010 passive sampler station	0.5 hours after beginning of rain
stormwater	5/13/2013	15:15	suspect culvert	1 hour after end of heavy rain
	6/12/2013	15:00	suspect culvert	0.5 hours into rain preceded by 12 days of mostly dry weather (0.03 cm on 6/11)
	6/12/2013	15:08	downstream culvert	
sediment traps	5/2/2013 - 6/20/2013	NA	14' downstream of I-2C	26 dry days, 24 days with rain, total = 8.4 cm
		NA	21' downstream of I-2C	
groundwater	4/25/2013	15:00	adjacent to I-2B	dry; no rain in 2 days
	4/26/2013	14:25	baseflow from suspect culvert	dry; no rain in 3 days
	4/26/2013	15:35	30' upstream of I-2B	

Sampling Details

Descriptions and photos of the various sampling techniques, stream measurements, and laboratory analyses are contained in Appendix C.

In-Situ Bioassays

Trout Toxicity Testing

Environment Canada (1998) developed a toxicity test using the embryo, alevin, and fry (EAF) lifestages of salmonids. Each lifestage is sensitive to different pollutants. An environmental exposure encompassing all of these lifestages is a true chronic test. The biological effects assessed can include mortality, failure to hatch, abnormal development, and reduced growth. The EAF early lifestage test works equally well in a laboratory or in hatchboxes set in a stream.

Rainbow trout in-situ testing for the study was conducted by Nautilus Environmental (Nautilus) with assistance from the Washington State Department of Ecology (Ecology). Nautilus used a method based on the British Columbia Ministry of the Environment *Field Sampling Manual* (BC MoE, 2003).

Nautilus obtained trout eyed-embryos for the *in-situ* toxicity testing from Trout Lodge in Sumner, Washington. Ecology acquired Hydraulic Project Approval (HPA), fish transport, and fish stock permits prior to deployment.

Two placement types were used for the trout tests. For stations I-1, I-2B, and I-2C, the standard in-substrate burial method was used (Figure 5). Due to the silty substrate at station I-2A, the crate method was used (Figure 6). Both the in-substrate and crate placement methods used Whitlock-Vibert hatchboxes. Thirty eyed-embryos were placed in each hatchbox. Three hatchboxes were deployed at each station for a total of 90 embryos per station.

In-substrate placement – method description

The hatchboxes were placed inside steel wire cages (approximately 7 by 14 inches). Washed stream gravel (1 to 2 inch diameter) was used to supplement the native stream gravel surrounding the hatchboxes and to hold them in place inside the cages. The cages were then zip-tied to keep them closed. See Figure 5 for a diagram and photograph of the arrangement of the cage placements in the stream.

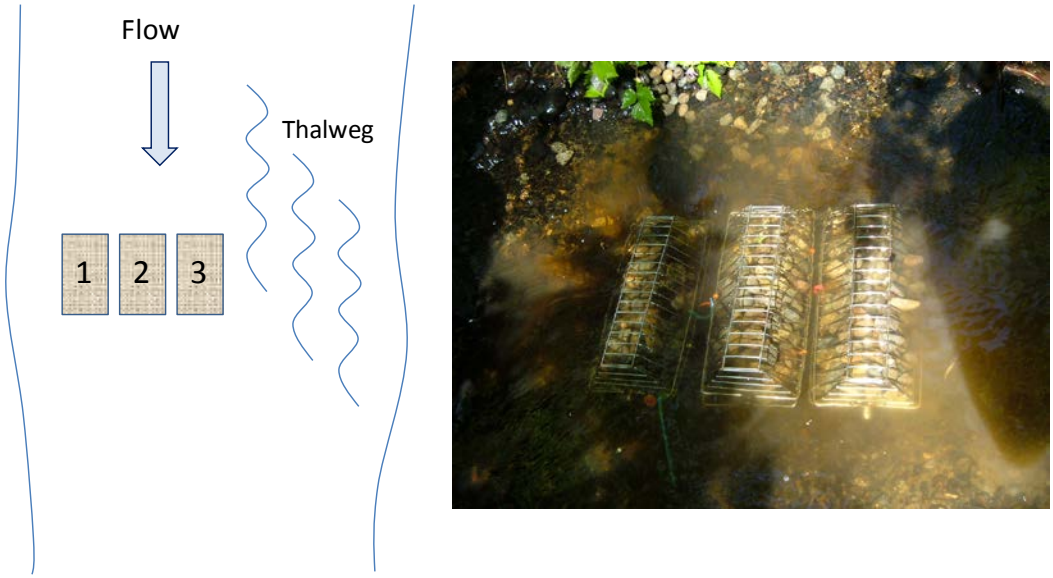

Figure 5. Diagram (left) and photo (right) of hatchbox in-substrate deployment method.

Field staff selected stream locations that had a steady unidirectional flow outside of the main current (thalweg). Excavations were dug at these locations deep enough so the tops of the cages would be at about the same elevation as the stream bed. The three cages were covered with a small mound of gravel after being placed side-by-side in the excavation at each station. A continuous temperature logger was deployed on one cage at each station.

Crate placement – method description

Plastic mesh sacks were placed inside of PVC milk crates. Hatchboxes were then placed inside the plastic mesh sacks. Washed stream gravel (1 to 2 inch diameter) surrounded the hatchboxes to hold them in place inside the mesh sack and milk crates. Bungee cords were used to keep the mesh sacks closed. The crates were then slid onto steel fence posts which kept the crates just above the substrate. The three crates were located equidistant across the stream since the velocities and depths were similar across the stream. A continuous temperature logger was deployed on one of the crates. Figure 6 is a photograph of the crates deployed at Station I-2A.

Figure 6. Photo of crate method for trout hatchbox deployment.

Eyed-embryos from the same batch of eggs from Trout Lodge were held at the Rainier Environmental Laboratory in Fife, Washington to assess the health of the batch of eggs. The lab trout were kept at a temperature close to the stream-exposed trout to track developmental milestones and time field visits for monitoring lifestage changes (alevin hatch and fry swim-up). The field checks involved removal, inspection, and reburial of the cages and hatchboxes. The number hatched, number alive, and observations on fish health were recorded at each field visit. Table 3 lists the dates and times of trout field activities.

Table 3. Timing of field activities for the trout.

Date	Time	Location	Survival	Lifestage	Action
4/30/2013	12:20	I-1	NA	eyed-embryo	3 replicates of 30 embryos deployed
	13:10	I-2A	NA	eyed-embryo	3 replicates of 30 embryos deployed
	13:35	I-2B	NA	eyed-embryo	3 replicates of 30 embryos deployed
	14:10	I-2C	NA	eyed-embryo	3 replicates of 30 embryos deployed
5/10/2013	13:00	I-1	98%	hatching	counted & redeployed
	13:40	I-2A	89%	hatching	counted & redeployed
	14:05	I-2B	81%	hatching	counted & redeployed
	14:30	I-2C	87%	hatching	counted & redeployed
5/17/2013	12:45	I-1	90%	alevin	replicate 1 taken for chemical analysis
	12:00	I-2A	60%	alevin	replicate 1 taken for chemical analysis
	11:40	I-2B	4%	alevin	terminated - survivors sent for chemical analysis
	10:55	I-2C	24%	alevin	terminated - survivors sent for chemical analysis
5/30/2013	11:00	I-1	93%	swim-up	terminated - survivors sent for chemical analysis
	12:20	I-2A	35%	swim-up	terminated - survivors sent for chemical analysis

Because of low survival (4.4%) at station I-2B on May 17, 2013 (around the instream exposure halfway point), the stream exposure was terminated and survivors were frozen for later metals analysis. It was feared that no trout from I-2B would be left for analysis at the end of the full exposure time. Station I-2B is located just downstream of the stormwater culvert suspected of causing mortalities seen at I-2C in 2010. Alevins from I-2C were removed at the same time as from I-2B and frozen for metals analysis because I-2C was a little further downstream of the culvert and comparison to the I-2B metals results was important. Station I-2C alevin survival was only 24.4% at the time. Removing trout for analysis terminated the I-2B and I-2C instream exposures.

To provide for comparisons to metals results from stations I-2B and I-2C, trout from one replicate hatchbox were taken on May 17 from both the lab and stations I-1 and I-2A. This meant that just two replicate hatchboxes at stations I-1 and I-2A and three replicates from the lab were left to go the full-term of the test (until swim-up). Exposures are usually terminated when trout reach swim-up to avoid adverse effects related to malnutrition after complete utilization of the yolk.

The trout remaining on May 30 at the end of the instream exposure were transported to the Rainier Environmental Laboratory in Fife, Washington for enumeration of deformities and for length and weight measurements. The lab trout were also sacrificed by being placed in Perrier carbonated water at the same time. The lab trout received the same measurements. The results from the trout counts and measurements were analyzed using CETIS v1.8.0.4 (Tidepool Scientific, 2010). More detail on the trout toxicity tests is provided in the Nautilus report in Appendix B.

Trout Tissue Metals

Directly after the trout fry were sacrificed and measured at Rainier Laboratory, Ecology staff placed composites of fry into certified contaminant-free jars provided by Ecology's Manchester Environmental Laboratory (MEL) and transported them to Ecology Headquarters where they were frozen prior to being shipped to MEL for metals analysis. The trout alevins from day 17 were previously placed in contaminant-free jars and frozen.

Trout samples were later shipped in an iced cooler to MEL for metals analysis. Each composite sample consisted of 4 - 56 whole fish (Table 4). The fish were digested whole body as part of the analysis preparation method (EPA 3051). The tissue samples were analyzed for arsenic, cadmium, copper, lead, nickel, and zinc.

Table 4. Fish tissue composite sample information for metals analysis.

Station	Process Date	Life Stage	Number in Composite	Sample Weight (g)
Lab Rep 1	5/17/13	Alevin	30	4
I-1 Rep 1			29	3
I-2A Rep 1			18	2
I-2B (Rep 1-3)			4	< 1
I-2C (Rep 1-3)			22	2
Lab Rep 2	5/30/13	Fry	29	4.9
Lab Rep 3			30	5.5
I-1 (Rep 2 & 3)			56	9.6
I-2A (Rep 2 & 3)			21	3.5

Biological Assessments

As part of the Deschutes River Multi-Parameter Total Maximum Daily Load Effectiveness Monitoring pilot project, periphyton and macroinvertebrate data were collected from 2010 through 2013 (Collyard and Von Prause, 2009). The full report will be published in 2015. The two locations on Indian Creek where in-situ trout were deployed, and environmental samples taken, were included in this 2013 report.

Composite macroinvertebrate and diatom samples were collected from riffle areas using methods outlined in the Quality Assurance (QA) Project Plan (Era Miller, 2013). Because the 2010 and 2013 trout deployments and environmental sampling occurred in the spring, biological data were collected outside the normal index period (July 1 through October 15) established for Washington State (Plotnikoff and Wiseman, 2001).

A suite of mutual periphyton and macroinvertebrate metrics commonly used for assessing stream health and determining stressors were calculated for each sample (Bahls, 1993; Barbour et al., 1999; Porter el al., 2008; Van Dam et al., 1994). The metrics of primary interest include those associated with metals and sediment and those that infer stream health. These metrics have been evaluated for the ability to distinguish impairment and are recommended as the most likely to be useful in other regions of the United States (Barbour et al., 1999).

For this 2013 study, stormwater impacts to Indian Creek were assessed by comparing metrics from sites upstream and downstream of the stormwater discharge. Using the upstream metrics site as baseline, the direction of metric responses from upstream to downstream sites was determined. Metric responses were then compared to published predicted biological metric responses (Barbour et al., 1999). Coefficients of variation (CVs) were determined for metrics between sites and compared to CVs calculated from 10 duplicate samples collected during the study period across Western Washington (Bahls, 1993; Ecology, unpublished data).

Based on predicted metric responses to stress, the stream health and stressors influencing biological communities were inferred by totaling mutual metrics for upstream and downstream Indian Creek sampling sites. Greater weight was given to metric differences where CVs were greater than duplicate values.

Periphyton

Periphyton is a community of microbes, algae, and bacteria living on hard substrate such as rock, shells, and logs in aquatic environments. A common analysis of periphyton, including for this study, focuses on algae, specifically diatoms. Similar to benthic macroinvertebrate assessments, diatom community assessments are a key indicator of stream health.

Periphyton and benthic macroinvertebrates were collected before trout hatchboxes and sediment traps were installed to avoid disturbance from placement of these devices. Periphyton was collected from native substrates at the upper Indian Creek site (near trout station I-1) and at the lower Indian Creek site (near trout station I-2C) near the same calendar date as in 2010. Periphyton was also collected at a reference site for comparison to the Indian Creek sites. The reference site was located in an undeveloped area of Capitol Forest near Olympia.

Riffle areas within site reaches were targeted. Reach length was determined by multiplying the average bankfull width times 20 (Adams, 2010). Sampling points within riffles were identified by establishing a minimum of 2 equally spaced transects across each riffle for a total of 8 transects per reach. At each transect, the distance from left to right was estimated, and the bottom substrate was sampled so that half the sampling occurred in mid-channel (50% wetted width) and half were in the margins (25% and 75% wetted width).

Periphyton was sampled by removing rocks from sampling points. Before processing, rock surfaces were lightly rinsed with reverse osmosis/de-ionized (RO/DI) water to remove loosely bound sediment and macroinvertebrates. The surfaces of the rocks were then scraped with a stiff plastic brush to remove the loosely attached periphyton matrix. This material was composited in a plastic tray, rinsed into a 1-L acid-washed bottle using RO/DI water, and placed on ice. A minimum of 125 cm^2 was sampled at each sampling point.

Periphyton samples were prepared for chlorophyll-a analysis by filtering a 10 ml sub-sample through a 0.45 micron filter and storing in acetone in the dark. Samples were split and centrifuged for percent total solids, total metals, and percent total organic carbon (%TOC) analysis. Results of periphyton metal concentrations are expressed as mg of metal/kg wet weight (ww) and have not been corrected based on %TOC or chlorophyll-a concentrations.

Periphyton samples were sent to Rhithron Associates, Inc. for taxonomic identification. Periphyton metals and total solids were analyzed by Brooks Rand Labs in Seattle WA, and chlorophyll-a and %TOC were analyzed by Ecology's Manchester Environmental Laboratory.

Benthic Macroinvertebrates

D-Frame Kicknet Sampling

Instream benthic macroinvertebrates were collected from the native substrate at both the upper and lower Indian Creek sites. Macroinvertebrates were collected following Ecology's collection protocols as described in the Ecology publication: *Benthic Macroinvertebrate Biological Monitoring Protocols for Rivers and Streams: 2001 Revision* (Plotnikoff and Wiseman, 2001).

Eight biological samples were collected from riffle habitat in a reach: 2 samples were from each of 4 riffles. A variety of riffle habitats were chosen within the reach to ensure representativeness of the biological community. This sampling design maximizes the chance of collecting a large number of benthic macroinvertebrate taxa from a reach.

Macroinvertebrate samples were collected with a D-Frame 500-micrometer mesh kicknet. The base of the D-Frame kicknet encloses a one-square-foot area of substrate in front of the sampler. Larger cobble and gravels within the sampled area were scraped by hand and brushed softly, visually examined to ensure removal of all organisms, then discarded downstream of the sampler. Remaining substrate within the sampler was then thoroughly agitated to a depth of 2 to 3 inches (5 to 8 cm). In order to have enough sample for taxonomic identification and metals analysis, side-by-side duplicate kick samples were taken at the same time.

Net contents were then emptied into a rinse tub by inverting the net and gently pulling it inside out. Tub contents were poured into a U.S. Standard No. 35 sieve. The tub was rinsed into the sieve and examined to ensure all organisms have been removed. This procedure was repeated for each of the 8 sub-samples.

All of the sieve contents were placed in a sample bottle. Each bottle was filled about 2/3 full to allow room for an alcohol preservative (85% non-denatured ethanol). Sample bottles were then labeled and shipped to Rhithron Associates, Inc. for taxonomic identification and to Brooks Rand Labs for metals analysis.

Bug Bags

Bug bags worked well for the 2010 study (Marshall and Era-Miller, 2012), and the same bug bag method was used in 2013. This method was similar to one used by the state of Maine (Davies and Tsomides, 2002). For 2013, bug bags were placed only at the lower Indian Creek monitoring site.

The bags were made using 2-inch gravel stuffed inside square pieces of mesh fencing (with 1-inch square holes) held together at the edges with zip ties. Each bag was 12 x 18 inches in dimension (Figure 7). Three bug bags were distributed in a transect encompassing at least 2 riffles at the lower site. Distance between the bug bags was approximately 20 ft.

A. Bug bag dimensions
B. Bags at deployment
C. Retrieval of bags

Figure 7. Bug bag method of benthic macroinvertebrate collection.

Upon retrieval, the bug bags were gently scooped up from the substrate with a D-Frame kicknet and then transferred into a tub. The mesh bags were cut open allowing rocks, debris, and bugs to fall into the rinse tub. Tub contents were then sieved and placed into sample bottles, in the same way as was done for the instream benthic macroinvertebrate collection. Samples were shipped to Rhithron Associates, Inc. for taxonomic identification.

Metals Correlations with Survival

Survival rates for the alevin and fry lifestages were correlated with metals concentrations in fish tissue using the Pearson product-moment correlation coefficient (Pearson’s r) and the Spearman’s rank correlation coefficient calculations in Microsoft Excel 7. The timing of survival counts and tissue sampling was standardized according to the trout EAF method described above. Field staff removed hatchboxes for observation and sampling when the lab trout held at the same temperature as the stream reached 2 milestones: the beginning of hatch (alevins) and the beginning of swim-up (fry). The standardization of lifestage and observations provides for consistency between years. Correlation calculations for metals in tissue with fry survival included data from both 2010 and 2013.

Combined Toxicity Estimations

Estimating the toxicity of a complex mixture, such as a sample from an urban stream, cannot be done solely using the results from chemical analysis. The concentrations of the individual chemical constituents of the sample do not add up to a number which can be related to overall toxicity. The individual concentrations must be normalized first to some common standard of aquatic toxicity before adding them together. These normalized values are called *toxic units* and are calculated by dividing a substance's concentration by its LC_{50}[1] or some other estimation of toxic threshold. Toxic units can then be added to estimate overall toxicity. When toxic units are calculated by dividing a pollutant's water concentration by its water quality criterion (WQC), the result is best called a criterion unit (CU).

The toxic units approach has been used successfully with environmental samples. Wildhaber and Schmitt (1996) calculated CUs from a variety of pollutants (metals, inorganics, and organics) measured in Great Lakes sediment pore water and found the sums of CUs to generally agree with toxicity test results on sediment samples. Hickey and Golding (2002) assessed the combined toxicity of metals to stream macroinvertebrates by summing CUs calculated by dividing measured metals concentrations by their WQC. Allert et al. (2010) divided stream metals concentrations by their WQC and found that the sums correlated well with riffle crayfish density ($r = -0.95$) and carapace length ($r = -0.91$).

Kortenkamp et al. (2009) noted from an extensive review of the available literature on mixture toxicity that deviations from additivity (i.e., antagonism or synergism) are rare and mostly confined to mixtures with only a few compounds. They recommend dose addition as the default approach for assessing mixture toxicity rather than pursuing different modes of action for multiple mixture constituents. Warne and Hawker (1995) evaluated data from toxicity testing of 104 toxic mixtures composed of 182 different chemicals and found a tendency toward additivity (and away from antagonism and synergism) as the number of components in a mixture increased. Mixtures with less than or equal to 10 components produced more antagonism and synergism than mixtures with more components. Assuming additivity for the combined toxicity of multiple pollutants in stormwater seems to be a reasonable simplification for screening purposes.

This approach was used for the Indian Creek metals data. It was also used to reassess the potential for combined PAH toxicity, since the PAH toxicity equivalency factors used with the 2010 monitoring results (Marshall and Era-Miller, 2012) are set to estimate total carcinogenicity and not predict risk to aquatic life. The water quality standards used to calculate CUs for metals and PAHs are set for protection of both fish and invertebrate aquatic life.

Estimation of the Combined Toxicity of Metals

The concentration of each metal measured in stream samples was divided by its Washington State water quality criterion found in WAC 173-201A-240. The copper, zinc, lead, and nickel criteria are calculated based on hardness. The acute CUs were the highest measured concentration for each metal divided by its acute criterion (criteria maximum concentration or

[1] Lethal Concentration 50 is the concentration of a chemical which kills 50% of a sample population.

CMC). The chronic CUs were the median of all of the measured concentrations for each metal divided by the chronic criterion (criterion continuous concentration or CCC). The resulting CUs were then summed to estimate potential combined effects. If simple additivity is assumed for combining the toxicity of individual metals, then CU sums ≥ 1 have the same potential to adversely affect water quality as a CMC or CCC exceedance by a single metal.

Estimation of the Combined Toxicity of PAHs

There are no state or federal water quality criteria to protect aquatic life from PAHs. However, the Netherlands (Verbruggen, 2012) has set maximum acceptable concentrations (MACs) for the protection of aquatic life. Because MACs are set solely to protect aquatic life from short-term concentration peaks (as happens with stormwater discharges), MACs were used to calculate PAH CU sums based on Indian Creek monitoring results. MACs are appropriate for screening PAH results from Indian Creek to see if PAHs may have contributed to adverse effects seen in the test trout or benthic organisms.

The concentration of each PAH was divided by its MAC and the resulting CUs were then summed. If simple additivity is assumed for combining the toxicity of individual PAHs, then CU sums ≥ 1 have the potential to adversely affect water quality the same as an exceedance for a single PAH. CU sums over 10 CUs indicate that safety margins in setting the MAC might be exceeded.

Baas and Kooijman (2010) used in-situ deployment of daphnids in Netherlands streams to find out if the national environmental standards (maximum permissible concentrations or MPCs) were protective under realistic environmental exposures. In some cases where no MPC was exceeded, all daphnids died within 30 hours. Baas and Kooijman (2010) concluded that mixtures of chemicals which individually were all below their MPC caused the mortalities. MPCs and the MACs presented in Table 14 are similar except that MPCs include consideration of human health effects and are in some cases lower than the MAC.

Data Quality

All data for the 2013 study were reviewed by the report authors, Manchester Laboratory and the contract laboratories. All data were found to meet the data quality objectives outlined in the QA Project Plan for the project (Era-Miller and Marshall, 2013). Some of the project data have been qualified due to concerns with data quality but are acceptable as qualified and reported. A detailed discussion of data quality for this project is available in Appendix D.

Results

Trout In-Situ Toxicity

Trout survival approximately halfway through stream deployment was similar in 2010 and 2013. Figure 8 shows 17-day survival from 2013, and Figure 9 shows 23-day survival from 2010 ("2nd Survival Rate" is based on survival counts from the second field visit after deployment). Mean survival at station I-2C was 48% on day 23 in 2010 and 24% on day 17 in 2013. The survival data at I-2C overlapped enough between the 2 years for the difference to not be significant. I-1 had 92% survival on day 23 in 2010 and 90% survival on day 17 in 2013.

The red dot in the box plots shows the mean survival. The edge of the light blue area furthest from the mean (red dot) is the median. The extent of light blue shows the degree of skew and departure from normality. The box plot itself shows the full range of the data.

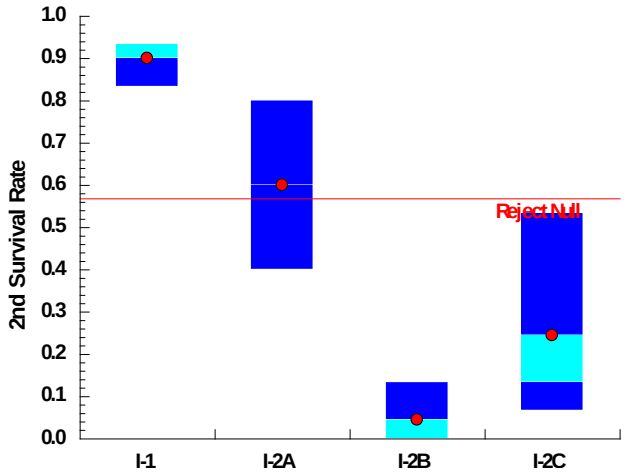

Figure 8. Trout survival from May 17 (day 17) in 2013.

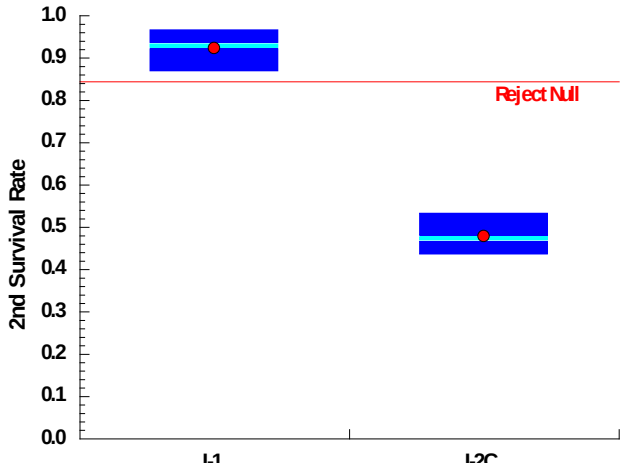

Figure 9. Trout survival from May 13 (day 23) in 2010.

Because of low survival (4%) at station I-2B on day 17 in 2013, the stream exposure was terminated for this station and the survivors sent for metals analysis. I-2B is just downstream of the stormwater culvert suspected of causing the mortalities seen at I-2C in 2010. Trout from I-2C were removed for metals analysis at the same time as those from I-2B because trout at these stations were exposed at different distances downstream of the culvert and comparison between tissue metals results would likely be important. Therefore, I-2B and I-2C have no survival results beyond 17 days exposure. Exposure for 17 days at I-2A, I-2B, and I-2C was sufficient for the results to pinpoint the location of the culvert between I-2A and I-2B as a source of toxicity.

Figure 10 shows that final survival (day 30) in 2013 at station I-2A was only 35%. I-2A is just upstream of the suspect culvert. Final survival in 2013 at I-1 was 93%. One or more pollutant sources are likely located between I-1 and I-2A. One possibility is the facility treating interstate highway (I-5) runoff for discharge to Indian Creek (see Figure 1). Day 30 survival is higher at I-1 than day 17 survival because of the removal of replicate 1 for metals analysis just after counting on day 17. Replicate 1 had the most mortality at that time.

Data reports for the trout in-situ toxicity test results are provided in Appendix B.

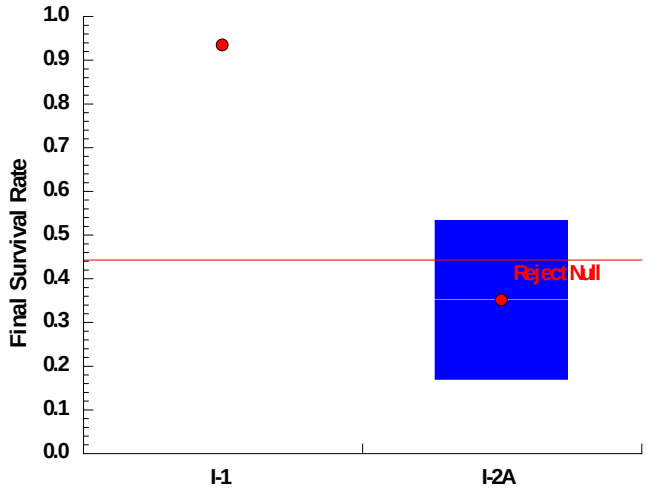

Figure 10. Final survival on May 30 (day 30) in 2013.

Metals in Trout Tissue

Results for the metals analysis of whole-fish composite samples from Indian Creek monitoring stations and from the laboratory trout are shown in Table 5.

Table 5. Whole body metals concentrations (mg/Kg, wet weight) in trout.

Sample ID	Lab		I-1		I2-A		I2-B		I2-C		Lab		Lab rep		I-1		I2-A	
Sample No.	1306038-01		-02		-03		-04		-05		-06		-07		-08		-09	
Lifestage	Alevins (4/30/13 - 5/17/13)										Swim-up Fry (4/30/13 - 5/30/13)							
Arsenic	**1.28**		**0.053**		**0.054**		0.278	U	**0.051**		0.049	U	0.046	U	0.050	U	0.048	U
Cadmium	**0.089**		0.048	U	0.049	U	0.278	U	0.050	U	0.049	U	0.046	U	0.050	U	0.048	U
Copper	**1.13**		**0.836**		**0.827**		**2.07**		**0.831**		**1.05**		**0.620**		**0.698**		**0.806**	
Nickel	**1.32**		**0.277**		**0.287**		**0.411**		**0.182**		**12.3**		**0.093**		**0.122**		**0.282**	
Lead	**0.323**		0.100	U	0.100	U	0.278	U	0.100	U	0.100	U	0.100	U	0.100	U	0.100	U
Zinc	**16.9**		**16.3**		**18.7**		**24.6**		**18.8**		**22.5**		**12.5**		**15.8**		**17.4**	

Bold values represent detected results.
U: not detected at or above the reported concentration.

Table 6 shows the correlation between alevin (day 17 in 2013) survival and copper, nickel, and zinc concentrations in fish tissue. Based on Pearson's r, copper had a moderate negative correlation with alevin survival. Zinc had a stronger negative correlation with alevin survival using both the parametric Pearson's r and the nonparametric Spearman's rank correlation.

Table 6. Metals in alevin tissue (mg/Kg, wet weight) and correlations with survival.

Station	I-1	I2-A	I2-B	I2-C	Pearson's r	Spearman's rank correlation
Date	5/17/13	5/17/13	5/17/13	5/17/13		
Copper (Cu)	0.836	0.827	2.07	0.831	-0.71	-0.40
Nickel (Ni)	0.277	0.287	0.411	0.182	-0.31	-0.40
Zinc (Zn)	16.3	18.7	24.6	18.8	-0.87	-1.00
Survival	90%	60%	4%	24%		

Table 7 shows the correlation between fry survival and copper, nickel, and zinc tissue concentrations. Fry final survival from both 2010 (34 days) and 2013 (30 days) were included in the correlation calculations. The results show a very strong negative correlation between copper tissue concentration and fry survival using both Pearson's r and Spearman's rank correlation. Nickel had a moderately strong negative Spearman's rank correlation but not Pearson's r.

Table 7. Metals in fry tissue (mg/kg, wet weight) and correlations with survival.

Station	I-1	I-1	I2-A	I2-C	Pearson's r	Spearman's rank correlation
Date	5/30/13	5/24/10	5/30/13	5/24/10		
Copper (Cu)	0.698	0.72	0.806	0.86	-0.99	-1.00
Nickel (Ni)	0.122	3.37	0.282	9.27	-0.60	-0.80
Zinc (Zn)	15.8	15.4	17.4	14.3	0.11	0.40
Survival	93%	89%	35%	14%		

Benthic Macroinvertebrate and Periphyton

Periphyton and macroinvertebrate metrics used in this analysis are presented in Appendix E, Tables E-1 and E-2. Metric results and calculated coefficients of variation (CVs) are reported in Appendix E, Tables E-3 through E-4.

The total numbers of stress-indicating metrics for macroinvertebrates and periphyton diatoms were calculated for upstream and downstream Indian Creek sampling sites. Table 8 presents all metrics evaluated while Table 9 presents only those metrics that had CVs greater than published pooled duplicate values.

Table 8. Totals for stream health, sediment quality, and metals exposure metrics.

Method	Overall stream health		Sediment		Metals	
	Upstream	Downstream	Upstream	Downstream	Upstream	Downstream
Diatoms	1	3	0	2	0	4
D-net	5	6	1	1	0	1
Totals	6	9	1	3	0	5

Table 9. Totals of the more significant metrics based on CVs > published values.

Method	Overall stream health		Sediment		Metals	
	Upstream	Downstream	Upstream	Downstream	Upstream	Downstream
Diatoms	0	0	0	1	0	3
D-net	3	3	0	0	0	0
Totals	3	3	0	1	0	3

Using upstream Indian Creek as a baseline, a total of 17 metrics out of the 24 assessed responded in a direction that indicates increased stress at the downstream site (Table 8). A total of 7 metrics out of the 10 assessed where CVs were greater than published pooled duplicate values also indicate increased stress at the downstream site (Table 9). Also, all metal indicator metrics increased at the downstream station, suggesting metals may be responsible for degrading conditions.

The periphyton metrics that did not predict increased stress at the downstream site included the pollution index, percent motile taxa, percent siltation taxa and percent motile taxa (Appendix E, Table E-1). The increase in the percent motile and siltation taxa suggests that sediment is impacting the periphyton community at the upstream site. The higher pollution index suggests the number of pollution-tolerant species of periphyton is higher at the upstream site.

All metrics describing general macroinvertebrate richness, with the exception of percent Ephemeroptera, Plecoptera, and Trichoptera (EPT), did not respond as predicted. A greater number of EPT taxa was observed at the downstream sampling station. However, the upstream station has higher overall percentages of these taxa (% EPT) than downstream (Table E-1).

Benthic Index of Biotic Integrity (BIBI)

Macroinvertebrate community data from upstream and downstream stations were compared using Ecology's Puget Sound Lowland BIBI (Wiseman, 2003) (Figure 11). The mean BIBI score from 2010-2013 was slightly higher at station I-1 when compared to I-2; however, CVs for BIBIs between stations was less than CVs of duplicate samples (Table E-4). This means that no differences between BIBIs where observed.

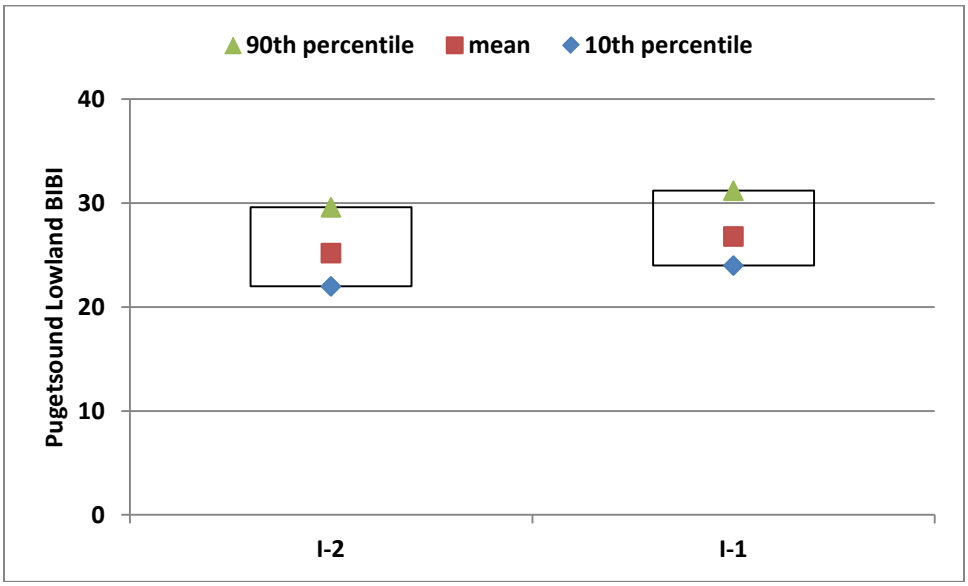

Figure 11. Pooled mean BIBI results from Indian Creek D-net samples (2010-2013).

2013 BIBI results from macroinvertebrate samples collected using bug bags at I-2 were slightly higher when compared to 2013 results using a D-net (Table 10 and Table E-5). The main metric responsible for the increase in BIBI score observed using bug bags was because of a higher number of taxa. An additional 8 species of *Chironomidae* were observed using the bug bags.

Table 10. 2013 metric values for the BIBI in lower Indian Creek using D-net and bug bags.

Metric Values	D-net	Bug bags
Taxa Richness	40	48
Ephemeroptera Richness	5	4
Plecoptera Richness	2	4
Trichoptera Richness	5	2
Pollution Sensitive Richness	1	2
Clinger Richness	10	11
Semivoltine Richness	3	4
Pollution Tolerant Percent	0.20%	2.60%
Predator Percent	6.47%	4.65%
Dominant Taxa (3) Percent	68.82%	56.32%
BIBI	24	26

Metals in Periphyton, Benthic Invertebrates, and Sediment Samples

Periphyton metals analysis produced a meaningful concentration gradient (I-2 > I-1 > reference stream) for arsenic, cadmium, copper, iron, lead, and zinc. Only manganese showed a similar gradient in the benthic macroinvertebrate results. There are no environmental standards against which to compare the periphyton or invertebrate tissue metals results. See the shades of green used to illustrate concentration gradients in Table 11.

Table 11. Metals results from instream biota.

Matrix:	Periphyton mg/kg ww			Inverts (mg/kg ww)		
Metal	ref	I-1	I-2	ref	I-1	I-2
Ag	0.0365	0.0375	0.071	U	U	U
Al	641.5	1615	1460	34.8	157	119
As	0.123	2.425	8.03	U	0.174	0.112
Cd	0.013	0.167	0.851	0.029	0.039	0.054
Cu	2.63	6.21	7.9	3.01	2.61	5.24
Fe	1185	8570	14700	48.3	962	539
Mn	49.95	12.65	18.05	9.45	107	176
Ni	4.925	3.97	4.52	0.35	1.19	0.92
Pb	0.785	59.6	295	0.03	0.454	0.309
Zn	3.725	2300	17500	32.7	65.8	43.5

Shade = increasing downstream gradient.

Table 12 shows that, except for nickel, metals concentrations in sediment were higher at station I-2 than I-1. The 2 sediment traps were set near the location of the I-2 bed sediment sample. The results from the 2 sediment traps (2 traps plus 1 replicate analysis) were in close agreement with each other and with the I-2 sediment sample. Suspended sediments and bottom sediments

were very similar in measured metals content. Full chemical results for the sediment trap samples are in Appendix E, Tables E-7 and E-8.

Table 12. Metals results from bottom sediments and sediment traps.

Matrix:	Sediment (mg/kg dw)		Sediment Traps (mg/kg dw)		
Metal	I-1	I-2	between I-2B and I-2C		
Ag	U	0.053	NA	NA	NA
Al	12000	14500	NA	NA	NA
As	3.16	4.38	5.16	5.55	4.84
Cd	0.137	0.266	0.28	0.28	0.28
Cu	12.3	24.3	22.4	21.5	20.1
Fe	17700	21500	NA	NA	NA
Mn	787	959	NA	NA	NA
Ni	28.3	26.6	21.9	22.4	22.1
Pb	11	21.9	20.1	21.1	19.9
Zn	73.8	137	151	155	147

Water Chemistry

Results for stream measurements and ancillary water chemistry parameters – hardness, alkalinity, TOC, dissolved organic carbon (DOC) and total suspended solids (TSS) – as well as chemistry for surface water, stormwater, and groundwater, are provided in Appendix E.

Total Risk from Metals or PAHs in Combination Based on Criterion Units

The sum of acute CUs based on the highest concentrations of metals measured in Indian Creek in 2010 (all except arsenic) and 2013 (all six metals) was 0.98 and very close to 1.0. Values of 1.0 and higher indicate the potential for the metals in combination to exceed (not meet) state water quality standards. The state has no water quality criterion for metals in combination. None of the measured metals concentrations exceeded its state water quality criterion nor did the sum of chronic CUs approach 1.0. See Table 13.

Table 13. Sum of criterion units for metals measured in downstream Indian Creek.

Metal:	Arsenic	Cadmium	Copper	Lead	Nickel	Zinc	Sum of CUs
Highest measured	3.55	0.025	7.96	0.242	1.56	12.2	
Criterion maximum concentration	360	2.05	10.19	35.52	892.77	72.14	**0.98**
Acute criterion units	**0.010**	**0.012**	**0.781**	**0.007**	**0.002**	**0.169**	
Median of measurements	0.74	0.012	1.24	0.184	0.80	3.71	
Criterion continuous concentration	190	0.64	6.54	1.24	91.14	60.54	**0.43**
Chronic criterion units	**0.004**	**0.019**	**0.190**	**0.148**	**0.009**	**0.061**	

CUs were calculated based on the list of PAH maximum acceptable concentrations (MACs) from the Netherlands. MACs are set to protect aquatic ecosystems from short-term exposure to concentration peaks (Verbruggen, 2012) and seem appropriate for an urban stream affected by stormwater. Table 14 shows that the 6/12/2013 stream sample exceeded 3 of the MACs for individual PAHs and produced a total of 11.7 CUs which exceeds the safety factor of 10 applied by the Netherlands in the development of MACs.

Table 14. Sum of criterion units for PAHs measured in downstream Indian Creek.

PAH	MAC	Upper culvert 6/12/13		Lower culvert 6/12/13		Stream sample 6/12/13		Stream sample 4/23/13	
		ug/L	**CU**	ug/L	**CU**	ug/L	**CU**	ug/L	**CU**
Methylated naphthalene	no MAC	0.003	NC	0.01	NC		NC		NC
2-Methylnaphthalene	no MAC	0.01	NC		NC		NC		NC
Acenaphthene	3.8	0.031	**0.008**		0	0.0069	**0.002**	0.0075	**0.002**
Acenaphthylene	33		0	0.085	**0.003**	0.033	**0.001**		0
Anthracene	0.10	0.018	**0.180**	0.017	**0.170**	0.011	**0.110**		0
Benzo(a)anthracene	0.10	0.054	**0.540**	0.023	**0.230**	0.024	**0.240**		0
Benzo(a)pyrene	0.010	0.087	**8.700**	0.039	**3.900**	***0.031***	**3.100**		0
Benzo(b)fluoranthene	no MAC	0.12	NC	0.058	NC	0.032	NC		NC
Benzo(g,h,i) perylene	0.0082	0.088	**10.732**	0.086	**10.488**	***0.039***	**4.756**		0
Benzo(k)fluoranthene	no MAC	0.08	NC	0.032	NC	0.024	NC		NC
Chrysene	0.070	0.11	**1.571**	0.094	**1.343**	0.039	**0.557**		0
Dibenz(a,h)anthracene	0.014		0		0		0		0
Fluoranthene	0.12	0.13	**1.083**	0.064	**0.533**	0.044	**0.367**		0
Fluorene	34	0.018	**0.001**		0		0		0
Indeno(1,2,3-cd) pyrene	no MAC	0.085	NC		NC		NC		NC
Naphthalene	130		0		0		0	0.015	**0.0001**
Phenanthrene	6.7	0.045	**0.007**	0.039	**0.006**	0.019	**0.003**		0
Pyrene	0.023	0.12	**5.217**	0.13	**5.652**	***0.058***	**2.522**		0
Retene	no MAC		NC	0.034	NC	0.015	NC		NC
∑CU			**28.0**		**22.3**		**11.7**		**0.002**

*** Red *italics*** = exceeds MAC

The pyrene and benzo(a)pyrene concentrations in the 6/12/2013 Indian Creek sample exceeded the Netherlands MAC. Pyrene and benzo(a)pyrene are products of incomplete combustion and common in urban stormwater. Benzo(g,h,i)perylene also exceeded its Netherlands MAC.

The stormwater CUs are provided solely for comparison. MACs are water quality standards that apply only to surface waters. The lower culvert in Table 14 refers to a culvert downstream of I-2C.

Oxygenated PAH (OPAH) Results

Table 15 shows results for the analysis of stream, stormwater, and groundwater samples for several OPAHs. The analysis for 3 of the OPAHs did not work, and the lab rejected (REJ) the results. Many of the reported concentrations are estimates and qualified (J) by the lab. The reported OPAH concentrations were similar in magnitude to the reported PAH concentrations in the study. Albinet et al. (2007) and Layshock et al. (2010) also observed PAH and OPAH concentrations to be similar in magnitude in environmental samples.

Table 15. Oxygenated PAH results for stream, stormwater, and groundwater.

Compound	Stream (ug/L)		Groundwater (ug/L)			Stormwater (ug/L)		
	I-2 4/23/13	I-2 6/12/13	Seep A 4/25/13	Seep C 4/26/13	Culvert Baseflow 4/26/13	Upper Culvert 5/13/13	Upper Culvert 6/12/13	Lower Culvert 6/12/13
9H-Fluoren-9-one	ND	ND	ND	ND	**0.052 J**	**0.062 J**	**0.064 J**	ND
Acenaphthenequinone	ND	**0.062**	ND	ND	ND	ND	**0.092**	ND
9,10-Anthracenedione	ND	**0.027**	ND	ND	ND	ND	**0.046**	ND
9,10-Phenanthrenedione	ND	REJ	ND	ND	ND	ND	REJ	REJ
1,4-Anthraquinone	ND	ND	ND	ND	ND	ND	ND	ND
Phenanthrene, 1, 4-dione	ND	REJ	ND	ND	ND	ND	REJ	REJ
4H-Cyclopenta(def)phenanthrene-4-one	ND	**0.013 J**	ND	ND	**0.017 J**	**0.017 J**	**0.017 J**	ND
Benzo(a)fluorenone	ND	**0.029**	ND	ND	ND	**0.020 J**	**0.055**	**0.069**
Benzanthrone	ND	ND	ND	ND	ND	**0.018 J**	ND	ND
Aceanthracenequinone	ND	ND	ND	ND	ND	ND	ND	ND
7,12-Benz[a]anthracenquinone	ND	ND	ND	ND	ND	ND	**0.069 J**	ND
Benzo[c]phenanthrene-1[1,4]quinone	ND	REJ	ND	ND	ND	ND	REJ	REJ
5,12-Naphthacenequinone	ND	ND	ND	ND	ND	ND	**0.076 J**	ND
Benzo[cd]pyrenone	ND	ND	ND	ND	ND	ND	**0.057 J**	**0.048 J**

Bold values indicate detected results
J = Analyte was positively identified; reported result is an approximate concentration
REJ = Result was rejected due to co-elution and was reported with another compound
ND = not detected

Water Temperature and Weather Data

Water Temperature

Water temperatures were not much different between the upper and lower sampling stations on Indian Creek. The lab trout were kept at a temperature as close to the stream temperature as possible. According to the Nautilus Environmental scientists conducting the trout testing, daily stream temperature changes were not sufficiently large or sudden enough to have adversely affected trout survival and development.

Weather at the time of stormwater sampling

Field staff sampled discharge from the suspect stormwater outfall (between station I-2A and station I-2B) 1 hour after the end of a rainstorm (1.5 hours after peak rain ended) on May 13, 2013. Stream temperature increased by 1.04° C at about the same time as the rainfall increased to peak intensity (0.28 cm/hour). There had been 0.05 cm of rain the day before and completely dry weather for 12 days preceding that very small rain.

Field staff sampled the suspect stormwater outfall again on June 12, 2013. The stormwater sample was taken 30 minutes after the rain began. A stormwater outfall downstream of all lower site stream activities was also sampled during this storm event. There had been 0.03 cm of rain the day before and completely dry weather for 11 days preceding that very small rain. A stream sample also was taken on June 12 about 50 minutes after the rain began. Unfortunately, Tidbits were only deployed along with trout, and both the trout and Tidbits had been removed from the stream by June 12. Ecology has no measurements of stream temperature for the second storm event.

Daily weather statistics for the 2013 study period are summarized in Appendix F.

Discussion

Historical Examples of Salmonid Mortalities in Regional Urban Streams

Maritime Heritage Fish Hatchery

In 1987, Ecology began an investigation of recurrent coho salmon (*Oncorhynchus kisutch*) mortalities at the Maritime Heritage Fish Hatchery in Bellingham, Washington (Kendra, 1988). Coho had been dying at the hatchery every autumn following the first or second significant rainfall at the end of the dry season. A kill also happened at the hatchery in spring of 1987 when a heavy rain fell after a dry spell. Hatchery losses typically ranged from 1% to 20%. Coho in Whatcom Creek died during the same rain events. Whatcom Creek is the water source for the hatchery.

No firm conclusions could be reached by the investigation into the coho deaths. No chemical was detected in samples of hatchery or creek water at concentrations known to be toxic. Gill lesions and a proliferation of chloride cells were found in dead fish and are an indication of environmental stress, perhaps from metals. The author hypothesized that synergistic metals toxicity may have caused the coho deaths. Sixteen of the 19 BNAs (base-neutral acid extractable organic compounds) detected in Whatcom Creek sediment were PAHs. Other than noting that PAHs originate in road runoff, the report did not discuss their potential role in coho mortalities.

The Ecology investigation lasted through 1990 and two more reports, Kendra and Willms (1990) and Ostergaard (1992), were written. Nothing much was discovered beyond the results of the initial study. A spring rain in 1990 killed 60% to 70% of coho at the hatchery, but sampling had been planned for the fall and was not ready at the time. Ostergaard (1992) noted that peak metals concentrations may have been missed in previous analyses by waiting for fish to respond before taking samples.

Prespawn Mortality

Feist et al. (2011) evaluated the relationship between land use and coho salmon prespawn mortality in the Puget Sound region. The strongest and most important relationships found were between prespawn mortality frequency and the extent of impervious surfaces in general, and roadways and commercial areas in particular. The common factor seems to be motor vehicles and related infrastructure.

Scholz et al. (2011) reported on the results of forensic investigations into the causes of coho prespawn mortality in Puget Sound area urban streams. A temporal relationship between coho mortality and rain storms was seen. Mortalities were worst early in the rainy season, especially if rains were delayed. Deaths seemed to be less following higher volume rainstorms. Mortalities decline after about a month of regular rainfall. Dead coho showed evidence of elevated exposure to both metals and PAHs. Coho showed no signs of exposure to pesticides or infectious disease.

Chemical Stressors

Metals

The strong negative correlation of copper in fish tissue with fry survival (Table 7) and moderate negative correlations for zinc and copper in tissue with alevin survival (Table 6) show there is a relationship between metals exposure and trout survival. This relationship might be cause and effect, or it might simply be covariation between copper and zinc and other environmental factors influencing trout survival.

Tissue concentrations are difficult to definitively relate to effects. Biegert and Valković (1980) compared tissue concentrations of copper, zinc, lead, and mercury to the mortality of rainbow trout exposed for 96 hours in a lab and could not find threshold tissue concentrations. Tissue levels were higher in survivors in some instances than in dead fish.

Neither copper nor zinc exceeded its water quality criterion in any stream sample during trout deployment in 2010 or 2013. The highest copper concentration in any stream sample was 78% of the criterion maximum concentration (CMC) calculated using the state's hardness-based approach but only 30% of the CMC calculated using EPA's biotic ligand model. The highest measured stream zinc concentration was only 17% of its CMC. The highest measured stream concentrations for the other four metals (arsenic, cadmium, lead and nickel) were all 1.2% or less of their CMCs. Using the criterion units (CU) approach shown in Table 13, the sum from all six metals was 0.98 CU and approached the threshold of 1.0 CU needed for a prediction of combined metals toxicity. Other metals may have been present in the samples but were not measured.

Sprague and Ramsey (1965) found that stronger mixtures ($\geq$ 2 times the incipient lethal level) of copper and zinc killed juvenile Atlantic salmon (*Salmo salar*) two to three times as fast as lethal concentrations of the metals individually. This could be an important consideration for urban streams which experience pulsed elevated concentrations of these two metals.

The highest copper and zinc concentrations measured in stream grab samples overlapped the range of toxic thresholds (LOECs and point estimates for 50% or lower effect levels) reported in EPA's ECOTOX database for diatoms exposed to copper or zinc. There was an overlap of 11% for freshwater diatoms exposed to copper and an overlap of 7% for freshwater diatoms exposed to zinc. Copper and zinc may have contributed to the periphyton effects seen in Indian Creek. None of the concentrations of the other metals measured for Indian Creek overlapped the range of diatom toxic thresholds reported in ECOTOX (http://cfpub.epa.gov/ecotox/).

Polycyclic Aromatic Hydrocarbons (PAHs)

PAHs are common pollutants in urban environments and come from (1) spillage of petroleum products (fuels or lubricants) or (2) combustion byproducts (Stein et al., 2006). Urban transportation provides an abundance of PAHs from both of these source categories along with the hard surfaces from which deposited PAHs can run into streams during precipitation events.

The sum of CUs calculated using the Netherlands PAH water quality criteria (maximum acceptable concentrations or MACs) and Indian Creek monitoring results was 11.7 CU (Table 14). This value is well over 1.0 CU and represents a risk for combined PAH toxicity.

The pyrene, benzo(a)pyrene, and benzo(g,h,i)perylene concentrations in the 6/12/2013 Indian Creek sample exceeded their Netherlands MACs. The Netherlands set MACs to protect aquatic ecosystems from short-term exposure to concentration peaks (Verbruggen, 2012). These exceedances are another example of the potential risk from PAHs to Indian Creek organisms.

However, none of the pyrene and benzo(a)pyrene concentrations measured in Indian Creek samples equaled or exceeded the thresholds for mortality found in EPA's ECOTOX database for fish, amphipods, daphnids, or insect larvae. The ECOTOX data included results from a 34-day exposure of rainbow trout early lifestages (Hannah et al., 1982).

Ankley et al. (1994) exposed an amphipod (*Hyalella azteca*), midge (*Chironomus tentans*), and oligochaete (*Lumbriculus variegatus*) to environmental samples of sediment contaminated with PAHs from an oil refinery. They found that toxicity to these test organisms correlated with the level of PAHs in the samples. In addition, follow-up exposures to UV light for two hours in clean water showed that photo-active substances had been bioaccumulated from the sediments sufficiently to quickly cause photo-induced toxicity. PAHs bioaccumulate in invertebrates. The sediment trap PAHs from Indian Creek (Appendix E, Table E-7) had PAH concentrations similar to all but the most contaminated samples from the Ankley study.

Substituted PAHs

Typical analyses for PAHs focus mostly on the original 16 EPA priority pollutant PAHs. None of the original 16 priority pollutant PAHs are substituted PAHs. Bornstein et al. (2014) tested various fractions of heavy fuel oil for toxicity to rainbow trout alevins (hatch to swim-up) and found that the alkylated PAHs were the most toxic fraction. Alkylated PAHs have one or more alkyl groups (alkanes) substituted onto the parent PAH ring structure. The original list of 16 PAHs is now commonly expanded in analysis to include two alkylated PAHs: 2-methylnaphthalene and retene (1-methyl-7-isopropyl phenanthrene). They are included in Table 14. There are many more alkylated PAHs of toxicological relevance, but they are rarely analyzed in environmental samples.

Barron et al. (2004) evaluated the results of oil exposure to pink salmon (*Oncorhynchus gorbuscha*) and Pacific herring (*Clupea pallasi*) embryos. Chemical analysis included 40 PAHs and alkylated homologs. The sum of toxic units calculated from the concentrations of the various alkylated phenanthrenes in eggs provided the best prediction of toxicity. The model was 80% accurate in predicting toxicity to herring embryos and 67% accurate in predicting toxicity to salmon embryos. The sum of criterion units in Table 14 is based on fewer PAHs than the Barron study and almost no alkylated homologs. The sum of criterion units in Table 14 would therefore tend to underpredict toxicity.

Because OPAHs are known to be toxic to fish (Knecht et al., 2013) and invertebrates (Lampi et al., 2005), our Ecology study included analysis of stream, stormwater, and groundwater samples for 14 OPAHs. Due to difficulties with co-elution, the analytical results are incomplete. The

OPAH analysis was an early attempt by Manchester Laboratory and inexperience affected the results. Layshock (2010) noted that analysis of OPAHs is in its infancy due to a limited number of authentic analytical standards and slow development of extraction and GC-MS procedures.

Mixtures of Metals and PAHs

According to Gauthier et al. (2014), the use of metals and fossil fuels drove industrialization and left widespread and ongoing contamination by both metals and PAHs. The authors report that mixtures of a small number of metals and PAHs resulted in synergistic toxicity in 44.7% of investigations of combined effects. The following mechanisms of metal-PAH synergism were among those proposed as contributors by the authors:

- PAH incorporation into the lipid layers of cell membranes increases permeability to metals by causing separation of cell membrane layers and loss of membrane integrity.
- Metals disrupt the cytochrome P450 system for metabolizing PAHs by down-regulating the expression of CYP1A1.
- PAHs inhibit metallothionein, thereby reducing the binding and removal of metals. Metallothionein is especially important for regulating copper and zinc.
- The above mechanisms create a positive feedback process whereby PAHs increase cellular metals which increase PAHs which increase metals and so forth.
- An abundance of reactive oxygen species (ROS) are created by ROS-active metals such as copper or cadmium combined with ROS-active PAHs such as phenanthrene or phenanthrenequinone.

The integrated ambient monitoring approach does not have the ability to detect synergism between metals and PAHs/OPAHs. However, the work by Gauthier et al. (2014) shows that synergism is a possibility. Using both metal and PAH sum of criterion units to screen for combined effects might partially account for such synergism.

Pesticides

Neither captan nor its breakdown product, tetrahydrophthalidimide (THPI), was detected in 2013 despite the large amount of captan in the 2010 results using a POCIS passive sampler (Marshall and Era-Miller, 2012). The 2010 detection in POCIS membrane extracts may have been spurious given that captan was also found in the trip blank and Manchester Laboratory listed it as tentatively identified in the analysis results for the POCIS extracts.

The Indian Creek BNA extractable results from 2013 found three herbicides and a breakdown product from one of these herbicides. These are not likely to have contributed to adverse effects to the trout deployed in the stream. King et al. (2013) exposed early lifestages of coho salmon (*Oncorhynchus kisutch*) to a mixture of the most common pesticides detected in urban streams in western Washington. The pesticide mixture included eight herbicides, two insecticides (carbaryl and diazinon), pentachlorophenol, and 4-nitrophenol. The authors concluded that exposure to the maximum concentration known to occur in western Washington streams did not pose a

significant risk for coho reproduction in urban streams. The coho early lifestages used in the study included the same lifestages as the rainbow trout exposed in Indian Creek.

Summary and Applicability of Monitoring Techniques

Trout Exposed to Streams

The trout in-situ method performed well in 2010 and 2013. The results at station I-1 were almost identical in both years. The results at I-2C were similar in both years. In 2013, stations I-2A and I-2B were not far apart in distance but had the suspect stormwater culvert between them. Trout mortalities at I-2B were much higher than at I-2A, pointing to the culvert as the source. The crate technique worked well at I-2A and showed that the approach is flexible enough to use in stream locations with substrate not suitable for burial of hatchboxes. The authors of this 2013 study feel that this technique is a reliable option in stream assessments for toxic contaminants.

Monitoring Metals in Periphyton (i.e., Benthic Biofilms)

Metals concentrations in periphyton showed spatial gradients (Table 11) that were consistent with known pollution sources and the biological impairment seen in the benthic community. The periphyton metals concentrations reported in Table 11 were higher than metals in macroinvertebrate tissue and better reflected the concentration gradients observed in stream water and sediments. Samples of periphyton are easy to collect for metals analysis. Periphyton is ubiquitous in surface water. Other studies have shown that periphyton metals analysis can be meaningful in assessing stream water quality:

- Rhea et al. (2006) discovered in biological and chemical monitoring results from the Boulder River watershed in Colorado that biofilm metals concentrations more frequently correlated with macroinvertebrate metrics than did the macroinvertebrate tissue metals concentrations. The authors propose that biofilm metals might make a good surrogate for metals in any compartment (water, sediment, or biota) of aquatic ecosystems.
- Ancion et al. (2010) exposed slides coated with a natural biofilm to simulated stormwater containing known concentrations of copper, lead, and zinc. The biofilm wet weight concentrations were enriched after 21 days exposure by up to 1500:1 for copper, 6000:1 for lead, and 500:1 for zinc. Community differences remained detectable for over 14 days after return to clean water.
- Ancion et al. (2013) demonstrated at 23 stream stations in New Zealand that measuring the metals content of stream biofilms was an ecologically relevant monitoring alternative to analyzing sediments for metals. The 23 stream sites included a mix of land use types ranging from forest to urban. Concentrations of copper, lead, and zinc in stream biofilms showed a linear correlation with sediment concentrations of the same metals. However, biofilm metals concentrations were usually higher and explained a greater proportion of bacterial and ciliate protozoan community variation than sediment metals concentrations.

Trout Exposed in the Laboratory

The trout held in the lab appeared to accumulate metals to the same or a higher degree than the stream-exposed trout. See Table 5. The lab trout were held at a hardness of 80 to 100 mg/L while the stream hardness ranged from 50 to 60 mg/L. The lower hardness of the stream would have encouraged metals bioavailability and concentration into fish tissue.

The relatively high concentrations of nickel in alevins and fry from the lab could be an indication of metals cross-contamination. We Ecology authors cannot know whether the lab trout were exposed to metals during incubation or whether the trout became cross-contaminated during preparation and shipment to Manchester Laboratory for analysis. Keeping biological samples free of nickel cross-contamination is difficult because of its prevalence in dust, hair, sweat, and saliva (Sunderman, 1993). The very high concentrations of nickel in trout tissue in 2010 (Marshall and Era-Miller, 2012) might also have been due to cross-contamination.

The lab trout were not intended to serve as negative controls but still should not have been exposed to metals to the degree they were. Because the lab trout had both good survival and relatively high reported tissue metals, using them in the calculation of correlation coefficients would have prevented finding the strong association of tissue copper with fry survival (Table 7) and of tissue zinc and copper with alevin survival (Table 6). The lab trout in 2010 were similarly high in measured tissue metals (Marshall and Era-Miller, 2012). Tissue metals in the 2010 lab trout could have confounded the analysis of the trout microarray results unless solely due to cross-contamination during handling after being sacrificed.

If the tissue metals measurements from the lab trout were not primarily due to cross-contamination, the high survival rate of the lab trout suggests that other chemicals (e.g., PAHs) may have been needed to cause deaths comparable to the stream-exposed trout. The lab and its equipment have many potential sources of metals but fewer sources for PAHs.

Assessments of Benthic Community Structure

Although results of bioassessments suggest the macroinvertebrate community is impaired at both monitoring locations (I-1 and I-2), an evaluation of macroinvertebrate and periphyton metrics suggest the causes of the impairment may be different. The evaluation of metric responses suggests that habitat (i.e., sediment) might be driving biological community structure at I-1, while water quality (i.e., metals) may be the biggest driver affecting biological communities below the suspect stormwater outfall. This conclusion is supported by results of metals concentrations in sediment, periphyton, and macroinvertebrate samples collected in 2013.

The results of BIBI scores suggest bug-bag sampling is comparable to results using D-nets. The differences between metric results are due to differences between sampling habitats. Using bug bags removes the influence of habitat on community structure and provides greater consistency when comparing sampling sites with different habitats. The disadvantage of using bug bags for this type of assessment is the methodology is unable to measure the effects of stormwater on habitat diversity. Additional replication and habitat parameters would need to be collected in order to account for these effects using D-net samples.

Conclusions

Issues related to urban stormwater and effects on salmon have been ongoing for some time. A hatchery in Oregon lost 10,000 coho salmon eggs after a hard rain in February 2014. (See http://www.kmtr.com/news/local/Staff-says-Coho-salmon-eggs-killed-in-hatchery-by-toxic-runoff-246968081.html for details.) Moreover, documentation of similar losses dates back at least to the 1980s at the Maritime Heritage Fish Hatchery in Bellingham.

Ecology studies in Indian Creek during 2010 and 2013 demonstrated a suite of methods for assessing the suitability of a stream for salmon reproduction. These studies used a combination of approaches which included *in-situ* bioassays and characterization of the chemical concentrations of the stream ecosystem. We were able to locate the specific stormwater pipe associated with the worst adverse effects to aquatic life and instream communities. Study results suggest metals and PAHs as possible co-factors in causing the adverse effects to test trout deployed in Indian Creek. The scientific literature provides several examples of mixtures of metals or PAHs acting together in an additive or synergistic way to increase toxicity to fish or invertebrates. Metals and PAHs have synergistic toxicity when present together.

Our study included use of the sum of criterion units to screen for additive effects from metals or PAHs. The use of criterion units seemed helpful in understanding stream biological impairment, and the science literature contains other examples of its usefulness. Assessing the combined effects of PAHs in this way required using water quality standards from the Netherlands.

In both 2010 and 2013, the assessment techniques appeared to have the right sensitivity. The upper site did not show impairment of instream communities and had good trout survival. The upper site is clearly urban with an interstate highway, suburbs, and commercial areas nearby, but the site does have a dense riparian buffer (Figure 2). Most of the trout died and instream communities were impaired at the lower site, located in a commercial area with a larger percentage of impervious surfaces (Figure 3). Stations repeated in 2013 had results consistent with the 2010 results. The 2010 study conclusion that transportation-related pollutants in stormwater caused adverse effects was confirmed in 2013. This conclusion should be enough to guide future management actions.

The most important steps for controlling damage from stormwater are reducing discharge volumes and surge flows, removing suspended solids, and controlling sources of the metals, PAHs, and other pollutants not reduced by solids removal.

Once stormwater controls are in place, trout eggs should be deployed and benthic communities assessed to determine the adequacy of control efforts. Given that complex mixtures in stormwater interact in unpredictable ways, only biological monitoring can judge whether stormwater controls are adequate.

The integrated monitoring approach also would also be useful for establishing ecological baseline conditions early in the development of a watershed and monitoring changes as they occur. The effectiveness of best management practices (BMPs) and low impact development (LID) practices could be assessed in this way. If stream impairment is seen, problems can be traced to the source. Preventing stream degradation is likely to be less expensive than remediation and retrofits.

Recommendations

- The techniques used in this 2013 study accomplished the most basic goals of an integrated ambient monitoring approach and should be repeated:
  - In-situ trout deployments with analysis of metals in fish tissue conducted as soon as significant mortalities (if any) are observed.
  - Identification and enumeration of benthic invertebrate and periphyton taxa.
  - Analysis of periphyton samples for metals and possibly PAHs.
  - Add or move stations as needed to pinpoint pollutant sources.
  - Sample stream water, groundwater, or sediments as needed for analysis of metals, PAHs, or OPAHs.
  - Use sum of criterion units to screen for combined effects from metals or PAHs,
- Add sampling stations between stations I-1 and I-2A to determine the contribution of other sources to the trout mortalities seen at I-2A in 2013. The WSDOT stormwater treatment facility just downstream of I-1 should be a primary focus since the ultimate goal is to use instream monitoring to evaluate treatment effectiveness.
- Continue to build a track record for bug bags. Bug bags may be better suited for assessing the water quality impacts from stormwater. Consider a similar approach for sampling periphyton using plates (artificial media) deployed instream as a substrate for periphyton.
- Employ the integrated ambient monitoring approach in the fall. Late September into October would be good for both rain and antecedent dry periods. Traditional BIBI scoring could be used in the fall and replace upstream-to-downstream comparisons. Another *Oncorhynchus* species could be tried since rainbow trout are reluctant fall spawners.
- Better represent peak pollutant loading. An automatic sampler set to take a sample when stream temperature has risen close to a degree Celsius within a short time might work well. Rain picks up both heat and pollution from impervious surfaces, especially after antecedent dry periods with sunshine (Marshall and Era-Miller, 2012).
- Note the presence of any stormwater outfalls or groundwater seeps when interpreting instream monitoring results. If needed, future sampling could include stormwater or groundwater. Sampling could include sediment traps for characterizing suspended sediments associated with storms. PAHs strongly partition to suspended sediments.
- Attempt, as much as possible, to use clean metals techniques in handling the test trout. Given the metals seen in the lab exposed trout tissue in 2010 and 2013, it is especially important that the lab establish clean techniques:
  - Analyze for metals a portion of the eyed eggs received from the hatchery.
  - Consider another attempt to use trout microarrays as diagnostic tools.
  - Hold the lab trout at a hardness of 50 to 60 mg/L to match the stream hardness.

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Appendices

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Appendix A. Glossary, Acronyms, and Abbreviations

Glossary

Alevin: The salmonid lifestage between hatching from the egg and swimming up into the water column. Alevin are characterized by having a yolk from which they derive the nutrition needed to survival and grow.

Ambient: Surrounding environmental condition (for example, surrounding air temperature).

Baseflow: Groundwater discharge to a surface stream or river. The component of total streamflow that originates from direct groundwater discharges to a stream.

Benthic: Bottom-dwelling organisms.

Biofilm: (see Periphyton).

Biotic Ligand Model (BLM): The BLM predicts heavy metal toxicity after complexation with organic (dissolved organic carbon) and inorganic (e.g., hydroxides, chlorides, carbonate) ligands and allows for competition with alkali and alkaline earth metals for fish gill binding sites.

Base/Neutral Acid (BNA): Organic compounds that are extracted into an organic solvent and analyzed by gas chromatography

Chorion: The acellular envelope surrounding a fish egg. The chorion hardens after fertilization in order to serve as a barrier and thereby protect the developing embryo.

Clean Water Act: A federal act passed in 1972 that contains provisions to restore and maintain the quality of the nation's waters.

Conductivity: A measure of water's ability to conduct an electrical current. Conductivity is related to the concentration and charge of dissolved ions in water.

Dissolved Oxygen (DO): A measure of the amount of oxygen dissolved in water.

ECOTOX: EPA's ECOTOX database is freely accessible online (http://cfpub.epa.gov/ecotox/). The database contains published toxicity test results from hundreds of species and individual chemicals. ECOTOX identifies the reference for each test result, allowing further inquiry into its relevance to an ambient monitoring project.

EDTA: Ethylenediaminetetraacetic acid (EDTA) binds metals in solution and reduces their bioavailability and toxicity.

Embryo: The fish lifestage occurring inside the egg. The embryo stage is when tissues differentiate and organs and body structures form.

Exceeded Criteria: When concentrations of a contaminant are higher than (do not meet) standards such as the Washington State Surface Water Standards for toxics (WAC 173-201A-240).

Fry: The salmonid lifestage commencing with swimming up into the water column after the yolk has been completely consumed. Fry must find and catch prey to provide the nutrition needed for survival and growth.

Gas Chromatography: Used in chemical analysis to volatilize and separate organic chemicals from a mixture in preparation for identification and quantification of these chemicals.

Grab Sampling: A discrete sample from a single point in the water column or sediment surface.

Incipient Lethal Level: The point in a concentration-response curve where acute toxicity ceases. The incipient lethal level (ILL) is determined by plotting the LC_{50} versus exposure time and finding the point at which the slope transitions to zero (i.e. becomes asymptotic). The LC_{50} determined in this way to be the ILL is the toxicant concentration at which the 50% surviving test organisms can be expected to continue living.

In-Situ Toxicity Test: A toxicity test conducted by placing test organisms into a container which allows flow-through of water and then placing the container into the stream, lake, or marine water of interest. An in-situ toxicity test provides a realistic environmental exposure without completely sacrificing the controlled conditions of a laboratory test. In particular, an in-situ toxicity test involves test organisms with a known history (e.g., age, health, prior chemical exposure) which are confined to one location for the test period. Because a realistic environmental exposure accepts the possibility of great variability and complexity, establishing cause and effect can be a challenge.

LC_{50}: Lethal Concentration 50 is the concentration of a chemical which kills 50% of a sample population.

LOEC: The Lowest Observed Effects Concentration (LOEC) is the lowest concentration of a substance in a toxicity test having a statistically significant difference from a nontoxic control. The LOEC is an approximation of the toxic threshold for that substance. Because only the concentrations used in the toxicity test are available to be the LOEC, the closeness of the LOEC to the true toxic threshold depends on the number and distribution of the concentrations used in the toxicity test.

Macroinvertebrate: Organisms on or in the stream substrate that are visible with the naked eye.

Metric: Index or method.

NOEC: The No Observed Effects Concentration (NOEC) is the highest concentration of a substance in a toxicity test not having a statistically significant difference from a nontoxic control. The NOEC is an approximation of the safe concentration for that substance. Because only the concentrations used in the toxicity test are available to be the NOEC, the extent to which

the NOEC is lower than the true safe concentration depends on the number and distribution of the concentrations used in the toxicity test.

Nonpoint source: Unconfined and diffuse sources of contamination. Pollution that enters water from dispersed land-based or water-based activities. This includes, but is not limited to, atmospheric deposition, surface-water runoff from agricultural lands, urban areas, or forest lands, subsurface or underground sources, or discharges from boats or marine vessels not otherwise regulated under the National Pollutant Discharge Elimination System (NPDES) program.

Parameter: Water quality constituent being measured (analyte). A physical, chemical, or biological property whose values determine environmental characteristics or behavior.

Passive Sampler: Passive samplers are devices for sampling water or air that do not require human or mechanical (pump) assistance. Passive samplers also do not collect the medium (water or air) along with the pollutants. Because of these features, passive samplers can be deployed for longer exposure times and with less effort. Passive samplers absorb pollutants similarly to living organisms.

Periphyton: A biofilm consisting of a complex mixture of algae, cyanobacteria, heterotrophic microbes, and detritus that is attached to submerged surfaces.

pH: A measure of the acidity or alkalinity of water. A low pH value (0 to 7) indicates that an acidic condition is present, while a high pH (7 to 14) indicates a basic or alkaline condition. A pH of 7 is considered to be neutral. Since the pH scale is logarithmic, a water sample with a pH of 8 is ten times more basic than one with a pH of 7.

Point Estimate: Point estimates, such as the LC_{50}, IC_{25}, or EC_{15}, are derived from toxicity test results to represent the concentration of the toxic substance which would cause a percent reduction equal to the specified effect level. For example, the LC_{50} is usually described as the concentration predicted to cause 50% mortality in a population of the test organisms. The IC_{25} estimates the concentration which would cause a 25% reduction in growth or reproduction. A "point estimate" is not really a single number but a range within which there is 95% confidence that the true value occurs.

Pollution: Contamination or other alteration of the physical, chemical, or biological properties of any waters of the state. This includes change in temperature, taste, color, turbidity, or odor of the waters. It also includes discharge of any liquid, gaseous, solid, radioactive, or other substance into any waters of the state. This definition assumes that these changes will, or are likely to, create a nuisance or render such waters harmful, detrimental, or injurious to (1) public health, safety, or welfare, or (2) domestic, commercial, industrial, agricultural, recreational, or other legitimate beneficial uses, or (3) livestock, wild animals, birds, fish, or other aquatic life.

Reactive Oxygen Species (ROS): Chemical oxygen species such as superoxide, hydrogen peroxide, and hydroxyl radical. At low levels, these species may function in cell signaling processes. At higher levels, these species may damage cellular macromolecules (such as DNA and RNA) and participate in the death of cells.

Salmonid: Any fish that belong to the family *Salmonidae*. In other words, a salmonid is any species of salmon, trout, or char.

Stormwater: The portion of precipitation that does not naturally percolate into the ground or evaporate but instead runs off roads, pavement, and roofs during rainfall or snow melt. Stormwater can also come from hard or saturated grass surfaces such as lawns, pastures, playfields, and from gravel roads and parking lots.

Surface waters of the state: Lakes, rivers, ponds, streams, inland waters, salt waters, wetlands, and all other surface waters and water courses within the jurisdiction of Washington State.

Swim-up: Trout life stage that begins when the alevin (larval salmonid) has absorbed its yolk sac and begins to swim upward to emerge from the gravels where eggs were deposited. The swim-up stage is viewed as a distinct life stage because the air bladder is not yet inflated, and the fish are negatively buoyant. They struggle to swim upward toward the water surface, and then gulp air to fill the air bladder. The swim-up stage ends once the air bladder is filled, and the juveniles are referred to simply as 'fry.'

Thalweg: The deepest and fastest moving portion of flow in a stream.

Water Quality Criteria: The maximum concentration of a chemical determined by EPA to be safe for aquatic life under short-term (acute) exposure or long-term (chronic) exposure.

Watershed: A drainage area or basin in which all land and water areas drain or flow toward a central collector such as a stream, river, or lake at a lower elevation.

Whitlock-Vibert hatchbox: The Whitlock-Vibert hatchbox is patented by the Federation of Fly Fishers and was developed for incubating trout and salmon eggs in streams to which these fish were being stocked. The hatchboxes have an upper egg chamber for embryos with slots through which the alevins slip after hatching into a lower nursery chamber. Nautilus adds extra screen to the nursery chamber so the fry cannot exit. Normally fry exit the nursery chamber when they are ready for swim-up. See: www.fedflyfishers.org/Default.aspx?tabid=4384 for more information.

Acronyms and Abbreviations

BIBI	Benthic Invertebrate Index of Biological Integrity
BLM	Biotic Ligand Model
BNAs	Bases, neutrals, and acids
CU	Criterion unit
CV	Coefficient of variation
DOC	Dissolved organic carbon
dw	dry weight
EAF	Embryo, alevin, and fry
EAP	Environmental Assessment Program
Ecology	Washington State Department of Ecology

EIM	Environmental Information Management database
EPA	U.S. Environmental Protection Agency
GCMS	Gas Chromatography/Mass Spectroscopy
LC_{50}	Lethal Concentration 50 (See Glossary for more information.)
LCMS	Liquid Chromatography/Mass Spectroscopy
LOEC	Lowest Observed Effects Concentration (See Glossary for more information.)
MQO	Measurement quality objectives
Nautilus	Nautilus Environmental (trout embryo test laboratory)
NOEC	No Observed Effects Concentration (See Glossary for more information.)
PAH	Polycyclic aromatic hydrocarbons
QA	Quality assurance
QC	Quality control
Rhithron	Rhithron Associates, Inc. (Missoula, MT)
RPD	Relative percent difference
SOP	Standard operating procedures
SRM	Standard reference materials
TIC	Tentatively identified compound
TOC	Total organic carbon
TSS	Total suspended solids
WAC	Washington Administrative Code
WQC	Water quality criteria
WSDOT	Washington State Department of Transportation
ww	wet weight

Units of Measurement

cfs	cubic feet per second
kg	kilograms, a unit of mass equal to 1,000 grams.
mg/Kg	milligrams per kilogram (parts per million)
mg/L	milligrams per liter (parts per million)
mL	milliliters
mm	millimeters
ng/L	nanograms per liter (parts per trillion)
ug/L	micrograms per liter (parts per billion)
um	micrometer

Appendix B. Nautilus Data Report

Results for the Rainbow Trout Early Life Stages *In-situ* Bioassay – Final Report

The 53-page Nautilus report is available as a separate pdf, linked to this report on the Internet at: https://fortress.wa.gov/ecy/publications/SummaryPages/1403050.html

Appendix C. Sampling and Analysis Information

Sample containers, preservation methods, holding times, and analytical methods for all environmental matrices (water, tissue, and sediment) are presented in Tables C-1 and C-2.

Surface Water Samples and Stream Measurements

Surface water samples were collected by hand as simple grabs from mid-channel following the EAP[2] *Standard Operating Procedure for Sampling Pesticides in Surface Waters, Version 2.1* (Anderson, 2012). Streamflow in Indian Creek is small and well-mixed, so single grabs were deemed adequate to represent creek water.

Streamflow Monitoring

Flow was measured using a Marsh-McBirney flow meter and top-setting rod as described in the EAP *Standard Operating Procedure for Estimating Streamflow: Version 1.0* (Sullivan, 2007). Flow was measured only once at upper and lower Indian Creek on April 24, 2013 during baseflow conditions. Flow monitoring was kept to a minimum to lower the potential for causing disturbance to the instream bioassessment tests.

Hydrolab and Tidbit Data

A MiniSonde® was used to measure ambient stream temperature, pH, conductivity, and dissolved oxygen each time a project-related activity occurred at the monitoring sites (e.g., during water sampling and in-situ trout deployment, checks, and retrieval). The MiniSonde® was calibrated and operated following the EAP *Standard Operating Procedure for Hydrolab® DataSonde® and MiniSonde® Multiprobes, Version 1.0* (Swanson, 2007).

Tidbit temperature loggers were deployed at each in-situ trout monitoring location and logged on the half-hour.

Metals

Collection of water samples for metals analyses followed the EAP *Standard Operating Procedure (SOP) for the Collection and Field Processing of Metals Samples, Version 1.3* (Ward, 2007). Surface water and stormwater were analyzed for both total and dissolved metals, while the groundwater seep samples were analyzed for dissolved metals only.

Samples for dissolved metals were filtered and acidified in the field using pre-cleaned filters from Manchester Environmental Laboratory. Field filtering and acidification generally took place within 15 minutes of collection.

[2] Environmental Assessment Program (Department of Ecology)

Stormwater

Stormwater was sampled directly from 2 stormwater runoff pipes near the lower Indian Creek monitoring site:

- The suspect culvert (suspected of causing trout mortalities in 2010) draining a nearby parking lot.
- A stormwater pipe draining the Plum Street interchange with Interstate-5 that is downstream of all other Indian Creek study activities.

The goal was to catch a stormwater runoff event after at least a 4-day antecedent dry period during April or May 2013, while the trout were instream. The first storm was narrowly missed on May 13 and only the suspect culvert had enough flow to sample 1.5 hours after peak rainfall and 1 hour after rain ceased. The timing of a second storm was better on June 12, allowing for adequate flow and sampling at both the suspect culvert and the downstream stormwater pipe only 0.5 hours after rain began and close to the time of peak rainfall. The June 12 samples, however, were taken after the test trout were out of the stream.

Groundwater

Groundwater was sampled once during the project from 2 seeps and from the suspect culvert at baseflow conditions on April 25 and 26, 2013, prior to the instream bioassessment tests. Baseflow from the culvert was sampled directly into bottles. The first attempt to sample the groundwater seeps was with piezometers (shallow wells). The piezometers did not produce enough water to effectively sample.

A licensed hydrogeologist from EAP, Kirk Sinclair, conducted the installation, sampling, and removal of the piezometers following EAP *Standard Operating Procedure for Installing, Measuring, and Decommissioning Hand-driven In-water Piezometers – Version 1.1* (Sinclair and Pitz, 2010). Ecology has a Hydraulic Project Approval (HPA) (No. 114142-2) from the Washington Department of Fish and Wildlife for installation of piezometers.

The second attempt to sample the seeps worked well. Seeps were sampled directly at their outlet by creating a small pool of seep water around the seep outlet with gravel from the stream. Samples were collected through Silastic® tubing with a peristaltic pump. Samples for dissolved organic carbon (DOC) and dissolved metals were filtered through a QED QuickFilter® (model FF8100) filter attached to the tubing. New tubing was used for each sample. Deionized water from Manchester Laboratory was passed through the same tubing at Ecology Headquarters and served as blanks for the samples.

Water quality measurements were taken onsite with the same MiniSonde® Hydrolab that was used for surface water and stormwater. The MiniSonde® was attached to a flow cell, and water was pumped from the seep through the flow cell. Measurements were recorded and samples taken once the MiniSonde® showed 3 stable readings approximately 3 minutes apart.

Suspended Sediments

Suspended sediments were collected with Hamlin sediment traps. One trap each was deployed at 2 locations at the lower Indian Creek monitoring site to capture suspended sediments in the creek, while the trout were instream. A picture of the Hamlin sampler is shown in Figure C-1. The Hamlin sampler is constructed using 14-gage solid stainless steel and has 2 distinct chambers. The top piece or "tongue" deflects flowing water up the ramp and into the ¼-inch-wide slots where water can fall through into the upper chamber. Dimensions are 21.5L x 9.25W x 4H inches. The weight (approximately 25 lbs) is enough to withstand low flows such as those in Indian Creek without being secured to the stream bottom (Lubliner, 2012).

Figure C-1. Hamlin sediment trap for collection of suspended sediments.

Assembled (right), Upper Chamber (top left) and Lower Chamber (bottom left, with baffles, tray, and exit ports). Lubliner, 2012.

Traps were de-contaminated prior to deployment with sequential rinses of hot water and Liquinox detergent followed by a 10% nitric acid rinse and deionized water. The traps were dried in a clean chemical hood before and after being rinsed with acetone. The traps were then covered with aluminum foil until deployment in the stream.

Sediment traps were deployed for approximately 52 days, including the time when study trout were instream. Fine materials within the traps were scooped out with decontaminated stainless steel spoons into certified contaminant free 1-liter jars and homogenized. Sediment jars were placed on ice and brought back to Ecology Headquarters and centrifuged to remove excess water. Samples were shipped on ice in a cooler to Manchester Laboratory for analysis.

Sample Containers, Preservatives, and Holding Times

Table C-1. Containers, preservations, and holding times for project samples.

Parameter	Matrix	Container	Preservation	Holding Time
DOC	Water[†]	60 mL poly bottle; 0.45 um pore size filters	Filter in field with 0.45 um pore size filter; 1:1 HCl to pH<2; Cool to 6°C	28 days
TSS		1 L poly bottle	Cool to 6°C	7 days
Alkalinity		500 mL poly bottle – no headspace	Refrigerate, 6°C	14 days
Hardness		Taken from the total metals sample bottle	HNO3 to pH<2 by the lab within 24 hours of collection	6 months after preservation
As, Cd, Cu, Ni, Pb, and Zinc		500 mL HDPE bottle	Field filter for dissolved; HNO3 to pH<2 by the lab within 14 days of collection	6 months after preservation
PAHs		Certified 1 liter amber bottle w/Teflon lid liner	Refrigerate, 6°C	7 days
OPAHs		Certified 1 liter amber bottle w/Teflon lid liner	Refrigerate, 6°C	7 days
Captan & THPI + TICs		Certified 1 liter amber bottle w/Teflon lid liner	Refrigerate, 6°C	12 hours
BNAs + TICs		Certified 1 liter amber bottle w/Teflon lid liner	Refrigerate, 6°C	7 days
As, Cd, Cu, Ni, Pb and Zinc	Fish Tissue	Certified 4-oz glass jar w/Teflon lid liner	Refrigerate at 6°C; can store frozen at -18°C	6 months; 2 years frozen
% Solids	Sediment	2-oz glass jar	Cool to 6°C	7 days; 6 months frozen
TOC		2-oz glass jar	Cool to 6°C	14 days; 6 months frozen
As, Cd, Cu, Ni, Pb and Zinc		Certified 4-oz glass jar w/Teflon lid liner	Transport at 6°C; can store frozen at -18°C	6 months; 2 years frozen
BNAs + TICs		Certified 4-oz glass jar w/Teflon lid liner	Transport at 6°C; can store frozen at -18°C	14 days; 1 year frozen

[1] Information in table was adapted from MEL (2008).

[†] The water matrix includes surface water, groundwater seeps and stormwater. Captan, THPI , and TSS was only analyzed in surface water and stormwater. BNAs were analyzed only in groundwater.

TOC: total organic carbon; DOC: dissolved organic carbon; TSS: total suspended solids
As: arsenic; Cd: cadmium; Cu: copper; Ni: nickel; Pb: lead
PAHs: polycyclic aromatic hydrocarbons; OPAHs: oxygenated PAHs
THPI: tetrahydrophthalidimide
BNAs: bases, neutrals and acids
TICs: Tentatively Identified Compounds

Chemical Analyses

Table C-2. Analytical methods for water, fish tissue, and sediments.

Analysis	Matrix	Analytical Method
DOC	Water†	SM 5310B
TSS		SM 2540D
Alkalinity		EPA 310.2; SM 2320B
Hardness		EPA 200.7; SM
As, Cd, Cu, Ni, Pb, and Zn (Dissolved and Total)		EPA 200.8; SM
PAHs (SIM) and OPAHs		GCMS, EPA method (modified) SW 846 8270
Captan and THPI + TICs		
BNAs + TICs		
As, Cd, Cu, Ni, Pb, and Zn	Fish Tissue	Preparation Method EPA 3051; EPA 200.8; SM
TOC	Sediment	PSEP - TOC
% solids		SM 2540G
As, Cd, Cu, Ni, Pb, and Zn		EPA 200.8; SM
#2 Diesel and Lube Oil		NWTPH-Dx
BNAs + TICs		GCMS, EPA method (modified) SW 846 8270

TOC: total organic carbon
DOC: dissolved organic carbon
TSS: total suspended solids
As: arsenic
Cd: cadmium
Cu: copper
Ni: nickel
Pb: lead
Zn: zinc
PAHs: polycyclic aromatic hydrocarbons
OPAHs: oxygenated PAHs
THPI: tetrahydrophthalidimide
BNAs: bases, neutrals and acids
GCMS: Gas Chromatography/Mass Spectroscopy
SM: Standard Methods
PSEP: Puget Sound Estuary Protocols
TICs: Tentatively Identified Compounds
† The water matrix includes surface water, groundwater and stormwater.

Appendix D. Project Data Quality

Trout In-Situ Toxicity Testing

The laboratory trout met all test validity criteria in EPS 1/RM/28, Biological Test Method: Toxicity Tests Using Early Lifestages of Salmonid Fish (Rainbow Trout), and are described in more detail in the Nautilus Final Data Report (Appendix B).

When adopting the nickel-plated barbecue baskets as the standard wire cages for holding hatchboxes, Nautilus performed laboratory toxicity testing under a variety of conditions to verify that the nickel-plating did not contribute to trout toxic responses.

Benthic Macroinvertebrate and Periphyton

All Quality Assurance/Quality Control (QA/QC) acceptance limits were met for the benthic macroinvertebrate and periphyton testing, as explained in the case narratives provided by Rhithron.

QC procedures for taxonomic determinations of invertebrates involved checking accuracy, precision, and enumeration. One sample was randomly selected, and all organisms were re-identified and counted by an independent taxonomist. Taxa lists and enumerations were compared by calculating a Bray-Curtis similarity statistic for the selected sample (Bray and Curtis, 1957).

QC procedures for periphyton taxonomy involved the re-identification of diatoms and non-diatom algae from a randomly selected sample by independent taxonomists. Re-identifications of diatoms and non-diatom algae were made internally at Rhithron. Bray-Curtis similarity statistics were generated by comparing the original identifications with the re-identifications, and adjustments to taxonomy were made where appropriate.

Results of QC procedures for sub-sampling and taxonomy are given in Table D-1.

Table D-1. Quality control results for macroinvertebrates and periphyton.

Station	Biotic Group	Sample Method	Sorting Efficiency (%)	Bray-Curtis Similarity for Taxonomy and Enumeration (%)
Indian 2 (lower)	Macroinvertebrate	D-net	100.0	77.6
Indian 1 (upper)			99.8	--
Indian 2 (lower)		Bug Bag	--	97.1
Indian 1 (upper)			98.2	--
Indian 2 (lower)	Periphyton	Riffle	--	--
Indian 1 (upper)			--	85.4

Sorting efficiency averaged 99.2% for macroinvertebrate samples, taxonomic precision for identification and enumeration was 98.5% for the randomly selected macroinvertebrate QA sample, and data entry efficiency averaged 100% for the project. Taxonomic precision for identification and enumeration was 85.4% for the randomly selected periphyton QA sample. These similarity statistics fall within acceptable industry criteria (Stribling et al., 2003; L. Bahls, personal communication).

Chemical Analyses

Laboratory measurement quality objectives (MQOs) were included for the following data quality measurements: laboratory control samples (LCS), laboratory duplicates or LCS duplicates, matrix spike recoveries (MS), matrix spike duplicates (MSD), and surrogate chemical recoveries (organics analyses only). Results for the laboratory MQOs are shown in Table D-2. Other QC measurements included laboratory method blanks, equipment blanks, trip blanks, and field replicates.

Fish Tissue Metals

MQOs for LCS and MS/MSD samples were met for the fish tissue metals as indicated in Table D-2. No sample or LCS duplicates were conducted for the initial analysis of data.

Replicate samples (1306038-06 and -07) from the laboratory test trout indicated huge differences in concentrations for nickel, copper, and zinc (arsenic, cadmium, and lead were not detected in the replicate samples). This was a highly unexpected result, so Manchester Laboratory was asked to re-analyze the sample extracts. Table D-3 shows results for the initial analysis (1st run) and the re-analysis of the initial extracts (2nd run). The relative percent differences (RPDs) between the first and second analyses showed good precision with only 3 of 30 RPDs > 20%, indicating that the variability between the replicate samples was an indication of sample variability and not due to issues with the analysis.

Manchester Laboratory also re-extracted archive of the laboratory test trout samples (1306038-06 and -07) and analyzed them in duplicate (1306038-06 dup and -07 dup). The digestion method used for the re-extracted samples (3rd run) was EPA 3050 (hot block) versus EPA 3051 (microwave) used for the initial samples (1st and 2nd runs). Results between the initial analyses and the re-extracted analyses were vastly different for nickel, but similar for copper and zinc, indicating high variability in the fish tissue nickel data or that the different digestion methods can affect nickel concentrations. Duplicate analyses for the re-extracted (3rd run) samples generally indicated acceptable precision with only 1 of 6 RPDs > 20%.

Table D-2. Results for Laboratory Measurement Quality Objectives (MQOs).

Parameter	Matrix / Sampling Dates	LCS (% Recovery)	Pass?	Duplicate samples (RPD)	Pass?	MS (% Recovery)	Pass?	MSD (RPD)	Pass?	Surrogate Recoveries (% Recovery)	Pass?
DOC	Surface Water 4/23/13 & 6/12/13	80 – 120	Yes	≤20%	Yes	75 – 125	Yes	20%	NAF	NA	NA
TSS		80 – 120	Yes	≤20%	Yes	NA	NA	NA	NA	NA	NA
Alkalinity		80 – 120	Yes	≤20%	Yes	NA	NA	NA	NA	NA	NA
Hardness		85 – 115	Yes	≤20%	NAF	75 – 125	Yes	20%	Yes	NA	NA
As, Cd, Cu, Ni, Pb, & Zinc		85 –115	Yes	≤20%	NAF	75 –125	Yes	≤20%	Yes	NA	NA
PAHs		10 – 150	Yes	≤40%	Yes (LCS dups)	20 – 150	NAF	40%	NAF	10 –150	Mostly (d)
OPAHs		10 – 150	Some (j,k)	≤40%	Mostly - LCS dups (k)	20 – 150	NAF	40%	NAF	10 –150	Mostly (l,m)
Captan & THPI + TICs		40 – 170	Yes	≤40%	Yes (LCS dups)	10 – 215	NAF	40%	NAF	15 – 180	Yes
DOC	Ground-water	80 – 120	Yes	≤20%	Yes	75 – 125	Yes	20%	NAF	NA	NA
Alkalinity		80 – 120	Yes	≤20%	NAF	NA	NA	NA	NA	NA	NA
Hardness		85 – 115	Yes	≤20%	NAF	75 – 125	Yes	20%	Yes	NA	NA
As, Cd, Cu, Ni, Pb, & Zinc		85 –115	Yes	≤20%	NAF	75 –125	Yes	≤20%	Yes	NA	NA
PAHs		10 – 150	Yes	≤40%	Yes (LCS dups)	20 – 150	NAF	40%	NAF	10 –150	Mostly (d)
OPAHs		10 – 150	Mostly (n)	≤40%	Mostly - LCS dups (n)	20 – 150	NAF	40%	NAF	10 –150	Yes
BNAs + TICs		50 – 150	Mostly (a)	≤50%	Mostly (LCS dups)	50 – 150	Mostly (b)	40%	Mostly (b)	30 – 150	Mostly (c)
DOC	Storm-water 5/13/13 & 6/12/13	80 – 120	Yes	≤20%	Yes	75 – 125	Yes	20%	NAF	NA	NA
TSS		80 – 120	Yes	≤20%	Yes	NA	NA	NA	NA	NA	NA
Alkalinity		80 – 120	Yes	≤20%	Yes	NA	NA	NA	NA	NA	NA
Hardness		85 – 115	Yes	≤20%	NAF	75 – 125	Yes	20%	Yes	NA	NA
As, Cd, Cu, Ni, Pb, & Zinc		85 –115	Yes	≤20%	NAF	75 –125	Yes	≤20%	Yes	NA	NA

Parameter	Matrix / Sampling Dates	LCS (% Recovery)	Pass?	Duplicate samples (RPD)	Pass?	MS (% Recovery)	Pass?	MSD (RPD)	Pass?	Surrogate Recoveries (% Recovery)	Pass?
PAHs		10 – 150	Yes	≤40%	Yes (LCS dups)	20 – 150	NAF	40%	NAF	10 –150	Mostly (d)
OPAHs		10 – 150	Some (k,o)	≤40%	Mostly - LCS dups (o)	20 – 150	Some (p)	40%	Mostly (p)	10 –150	Mostly (m,q)
Captan & THPI + TICs		40 – 170	Yes	≤40%	NAF	10 – 215	Yes	40%	Yes	15 – 180	Yes
As, Cd, Cu, Ni, Pb, & Zinc	Fish Tissue	85 – 115	Yes	≤20%	NAF	75 – 125	Yes	20%	Yes	NA	NA
TOC	Sediment	80 – 120	Yes	≤20%	Yes	NA	NA	NA	NA	NA	NA
% solids		NA	Yes	≤20%	Yes	NA	NA	NA	NA	NA	NA
As, Cd, Cu, Ni, Pb, & Zinc		85 – 115	Yes	≤20%	NAF	75 – 125	NAF	20%	NAF	NA	NA
BNAs		50 – 150	Mostly (e)	≤50%	Mostly (f) LCS dups	50 – 150	Mostly (g)	40%	Mostly (h)	18 – 150	Mostly (i)

NA = Not applicable.
NAF = Not analyzed for.
LCS = Laboratory Control Sample
MS and MSD = Matrix Spike and Matrix Spike Duplicate
DOC = Dissolved Organic Carbon
TOC = Total Organic Carbon
TSS = Total Suspended Solids
BNA = Bases, neutrals, and acids analysis
PAH = Polycyclic Aromatic Hydrocarbons
OPAH = Oxygenated PAH
THPI = Tetrahydrophthalimide
TICs = Tentatively Identified Compounds
Yes = Defined as 100% of the specific QA/QC compounds were within acceptance limits defined by the laboratory measurement quality objectives (MQOs).
Mostly = Defined as >50% of the specific QA/QC compounds were within acceptance limits defined by the laboratory MQOs.
Some = Defined as <50% of the specific QA/QC compounds were within acceptance limits defined by the laboratory MQOs.

(a) LCS recoveries were within QA/QC acceptance limits except for Carbazole and 3-Nitroaniline. The associated results were qualified as estimates. The RPDs of some analytes exceeded limits, but these analytes were not detected in any of the samples, so no qualifications were needed. 97% (75/77) of LCS compounds met limits. 78% (60/77) of LCS compounds met duplicate RPD limits.

(b) Most MS/MSD compounds recovered within limits [95% (71/75) for MS and 93% (70/75) for MSD]. 81% (61/75) of MS/MSD RPDs met limits. See case narrative for full explanation of which compounds (2,4,5-Trichlorophenol and BEHP) affected qualification of sample results.

(c) Only 2 of 20 surrogate compounds were outside limits among the samples. Surrogate recoveries were 90 – 100% within limits. The 2 chemicals that recovered low were 4-Chloroaniline and 4-Methylphenol.

(d) Seven surrogate compounds were analyzed with PAH water analyses. For both storm events (5/13/13 and 6/12/13), surface water collected on 4/23/13, and for the groundwater samples, one compound Fluorene-D10 recovered low, and all samples results associated with this compound were qualified (6/7 or 86% of the compounds within acceptance limits). The stream sample taken on 6/12/13 had low recovery for 3 surrogate chemicals (Fluorene-D10, Benzo(a)pyrene-D12, and Pyrene-D10). All 3 associated chemicals for this sample were qualified accordingly.

(e) LCS recoveries were good except for 9 of 76 chemicals in the LCS and 10 of 76 (88% and 87% met limits) in the LCS duplicate. Associated results in the samples were either qualified with a "UJ" instead of a "U" or with an REJ when there was no recovery at all. REJs included Benzoic Acid, 2,4-Dinitrophenol, and Triethyl Citrate.

(f) RPDs were quite good with 92% (70/76) meeting QC limits. They were not calculated for the 3 rejected chemicals (Benzoic Acid, 2,4-Dinitrophenol, and Triethyl Citrate). RPDs were slightly high for caffeine and cholesterol, but since there were no detections in associated samples, there was no need for qualifications to the associated non-detected results.

(g) Matrix spikes had 80% (61/76) meeting QC limits, and the MSD had 86% (65/76). The source sample had to be diluted 1:10 for analysis which led to non-recovery of some compounds. 6 chemicals were not recovered at all in either the MS or MSD. 2-nitroaniline wasn't recovered in the MS. The chemicals were qualified as REJ in the associated field sample 1303040-01. Several other chemicals recovered either high or low and were qualified as UJ since there was no detection of these chemicals in the samples.

(h) MS and MSD RPDs were 91% within acceptance limits (69/76).

(i) Surrogates had 85% acceptance in most samples (17/20). Some samples did even better. Only 2 chemicals required qualification: 4-Chloroaniline and 4,6-Dinitro-2-methylphenol. 4-Chloroaniline was rejected in all samples and the blank due to no recovery. 4,6-Dinitro-2-methylphenol was rejected only in the blank due to non-recovery. Bis(2-Chloroethyl)Ether-D8 and Phenol-D5 recovered high in some samples, but because there were no detections, no qualification was needed.

(j) For surface water samples collected 4/23/13, 4 of 14 LCS compounds did not recover at all (71% within acceptance limits). All 4 associated results were rejected. No duplicate LCS for 4/23/13.

(k) For surface and stormwater samples collected 6/12/13, 8 of 14 LCS recoveries did not meet acceptance limits (only 43% met limits). Three compounds were not recovered at all, and associated results were rejected. The other 5 compounds had a mix of low and high recovery, and the associated results were qualified as estimates. Duplicate LCS RPDs were all within acceptance limits of ≤40% except for the 3 rejected compounds.

(l) Surrogates for the samples collected 4/23/14 recovered within acceptance limits.

(m) For surface and stormwater samples collected 6/12/13, 1 of the 2 surrogate compounds (9H-fluoren-9-one-D8) recovered high for most of the samples. Compound 9H-fluoren-9-one was qualified as an estimate for all the affected samples.

(n) For groundwater, 4 of 14 LCS compounds did not recover at all (71% within acceptance limits). All 4 associated results were rejected. Duplicate LCS RPDs were all within acceptance limits of ≤40% except for the 4 rejected compounds.

(o) For stormwater collected 5/13/13, 4 of 14 LCS compounds were outside limits (71% acceptance). Two chemicals did not recover at all and 2 chemicals recovered low. LCS duplicates were poor for 3 of the compounds. Results associated with all 4 chemicals were qualified.

(p) MS/MSDs were only analyzed with the 5/13/13 samples. Nine of 14 compounds were outside limits (31% acceptance). Six of 14 MS/MSD RPDs were outside limits (57% acceptance). Results associated with the compounds outside limits were qualified.

(q) For stormwater collected 5/13/13, the 2 surrogate compounds were within acceptance limits for all but the MS/MSD samples where compound 9H-flouren-9-one recovered high. This compound was qualified as an estimate in the associated result sample.

Table D-3. Precision of fish tissue metals data (mg/Kg, wet weight).

	Nickel					Copper					Zinc				
Sample No.	Ist Run	2nd run	RPD	3rd run	RPD	Ist Run	2nd run	RPD	3rd run	RPD	Ist Run	2nd run	RPD	3rd run	RPD
1306038-01	**1.32**	**0.896**	**38%**			**1.13**	**0.794**	**35%**			**16.9**	**15.5**	**9%**		
1306038-02	**0.277**	**0.306**	**10%**			**0.836**	**0.872**	**4%**			**16.3**	**16.8**	**3%**		
1306038-03	**0.287**	**0.322**	**12%**			**0.827**	**0.821**	**1%**			**18.7**	**19.3**	**3%**		
1306038-04	**0.411**	**0.467**	**13%**			**2.07**	**1.106**	**61%**			**24.6**	**25.3**	**3%**		
1306038-05	**0.182**	**0.211**	**15%**			**0.831**	**0.848**	**2%**			**18.8**	**20.1**	**7%**		
1306038-06	**12.3**	**12.826**	**4%**	**2.597**		**1.05**	**1.057**	**1%**	**0.881**		**22.5**	**23.5**	**4%**	**23.7**	
1306038-06 dup				**1.775**	**38%**				**1.091**	**21%**				**24.0**	**1%**
1306038-07	**0.093**	**0.089**	**5%**	**0.214**		**0.62**	**0.612**	**1%**	**0.545**		**12.5**	**13.2**	**5%**	**14.6**	
1306038-07 dup				**0.239**	**11%**				**0.593**	**8%**				**15.0**	**3%**
1306038-08	**0.122**	**0.124**	**2%**			**0.698**	**0.720**	**3%**			**15.8**	**17.2**	**9%**		
1306038-09	**0.282**	**0.317**	**12%**			**0.806**	**0.824**	**2%**			**17.4**	**18.3**	**5%**		

	Arsenic					Cadmium					Lead				
Sample No.	Ist Run	2nd run	RPD	3rd run	RPD	Ist Run	2nd run	RPD	3rd run	RPD	Ist Run	2nd run	RPD	3rd run	RPD
1306038-01	**1.28**	0.050 U	NC			**0.089**	0.05 U	NC			**0.323**	0.1 U	NC		
1306038-02	**0.053**	**0.053**	**0%**			0.048 U	0.048 U	NC			0.1 U	0.1 U	NC		
1306038-03	**0.054**	**0.060**	**11%**			0.049 U	0.049 U	NC			0.1 U	0.1 U	NC		
1306038-04	0.278 U	0.278 U	NC			0.278 U	0.278 U	NC			0.278 U	0.278 U	NC		
1306038-05	**0.051**	**0.053**	**4%**			0.050 U	0.050 U	NC			0.1 U	0.1 U	NC		
1306038-06	0.049 U	0.049 U	NC	0.08 U		0.049 U	0.049 U	NC	0.08 U		0.1 U	0.1 U	NC	0.08 U	
1306038-06 dup				0.08 U	NC				0.08 U	NC				0.08 U	NC
1306038-07	0.046 U	0.046 U	NC	0.08 U		0.046 U	0.046 U	NC	0.08 U		0.1 U	0.1 U	NC	0.08 U	
1306038-07 dup				0.08 U	NC				0.08 U	NC				0.08 U	NC
1306038-08	0.050 U	0.050 U	NC			0.050 U	0.050 U	NC			0.1 U	0.1 U	NC		
1306038-09	0.048 U	0.048 U	NC			0.048 U	0.048 U	NC			0.1 U	0.1 U	NC		

Bold values indicate detected results
U = Not detected above the reported quantitation limit
NC = Not calculated
RPD = Relative percent difference

The variability of the laboratory test trout replicate samples led the project authors to decide that the laboratory test trout data were not useful for comparison to the in-situ trout data.

Sediments

MQOs for the sediment trap data are shown in Table D-2. The BNA analysis had some results that were outside of QC limits. Results for these analyses were used as qualified by Manchester Laboratory.

Both a field replicate and a processing split were analyzed for the sediment trap samples at lower Indian Creek as listed in Table D-4. RPDs above 20% were only for some of the PAH results and are highlighted in gray. The fact that the processing split had higher variability than the field replicate sample points to the heterogeneity of the fluvial sediment matrix.

Surface Water

MQOs for surface water chemistry are shown in Table D-2. PAHs and OPAHs were the only parameters to have some results outside of QC limits and were used as qualified by Manchester Laboratory.

A trip blank was analyzed for the surface water and stormwater samples collected on 6/12/2013 (Appendix E, Tables E-9 and E-10). It contained no detections of any of the target chemicals. The trip blank was processed by transferring deionized water in clean bottles from the analytical laboratory (Manchester) into sample bottles in the field.

A field replicate was analyzed for the water sample (Sample ID I-2 SW) collected on 4/23/14 and is shown in Table D-4. RPDs are reported for detected chemicals only and range from 0 – 14%, representing good precision.

Stormwater

MQOs for stormwater chemistry are shown in Table D-2. PAHs and OPAHs were the only parameters to have some results outside of QC limits and were used as qualified by Manchester Laboratory.

A trip blank was analyzed for the stormwater sample collected on 6/12/2013 (Table E-10). The trip blank had no detections of any of the target chemicals. The trip blank was processed by transferring deionized water in clean bottles from the analytical laboratory (Manchester) into sample bottles in the field.

Table D-4. Precision for detected chemicals in field replicates and processing splits.

Parameter	UOM	Sample ID	QC Sample Type	Rep 1		Rep 2		RPD (%)
Surface Water								
Total Suspended Solids	mg/L	I-2 SW	Field Replicate	6		6		0%
DOC	mg/L	I-2 SW	Field Replicate	6.5		6.5		0%
Alkalinity	mg/L	I-2 SW	Field Replicate	49.1		49.3		0%
Hardness	mg/L	I-2 SW	Field Replicate	52.5		52.5		0%
Arsenic - total	ug/L	I-2 SW	Field Replicate	0.74		0.73		1%
Arsenic - dissolved	ug/L	I-2 SW	Field Replicate	0.62		0.62		0%
Copper - total	ug/L	I-2 SW	Field Replicate	1.12		1.16		4%
Copper - dissolved	ug/L	I-2 SW	Field Replicate	0.65		0.66		2%
Nickel - total	ug/L	I-2 SW	Field Replicate	1.32		1.32		0%
Nickel - dissolved	ug/L	I-2 SW	Field Replicate	1.09		1.1		1%
Lead - total	ug/L	I-2 SW	Field Replicate	0.52		0.53		2%
Lead - dissolved	ug/L	I-2 SW	Field Replicate	0.178		0.172		3%
Zinc -total	ug/L	I-2 SW	Field Replicate	5.2		5.5		6%
Zinc - dissolved	ug/L	I-2 SW	Field Replicate	2.9		2.9		0%
Naphthalene	ug/L	I-2 SW	Field Replicate	0.015		0.013		14%
Sediment								
% Solids	%	I-2 SED LWR	Processing Split	54.2		53.9		1%
% TOC	%	I-2 SED LWR	Processing Split	4.5		3.9		15%
Arsenic	mg/Kg dw	I-2 SED LWR	Processing Split	5.55		4.84		14%
Cadmium	mg/Kg dw	I-2 SED LWR	Processing Split	0.28		0.28		0%
Copper	mg/Kg dw	I-2 SED LWR	Processing Split	21.5		20.1		7%
Nickel	mg/Kg dw	I-2 SED LWR	Processing Split	22.4		22.1		1%
Lead	mg/Kg dw	I-2 SED LWR	Processing Split	21.1		19.9		6%
Zinc	mg/Kg dw	I-2 SED LWR	Processing Split	155		147		5%
Lube Oil	mg/Kg dw	I-2 SED LWR	Processing Split	760		920		19%
Benz[a]anthracene	ug/Kg dw	I-2 SED LWR	Processing Split	75	J	140	J	60%
Benzo(a)pyrene	ug/Kg dw	I-2 SED LWR	Processing Split	160	J	200	J	22%
Benzo(b)fluoranthene	ug/Kg dw	I-2 SED LWR	Processing Split	210	J	280		29%
Benzo(ghi)perylene	ug/Kg dw	I-2 SED LWR	Processing Split	310	J	310	J	0%
Benzo(k)fluoranthene	ug/Kg dw	I-2 SED LWR	Processing Split	120	J	160	J	29%
Chrysene	ug/Kg dw	I-2 SED LWR	Processing Split	160	J	220	J	32%
Fluoranthene	ug/Kg dw	I-2 SED LWR	Processing Split	220	J	340		43%
Indeno(1,2,3-cd)pyrene	ug/Kg dw	I-2 SED LWR	Processing Split	230	J	250	J	8%
Phenanthrene	ug/Kg dw	I-2 SED LWR	Processing Split	84	J	140	J	50%
Pyrene	ug/Kg dw	I-2 SED LWR	Processing Split	210	J	330		44%
% Solids	%	I-2 SED UP & LWR	Field Replicate	48.7		54.2		11%

Parameter	UOM	Sample ID	QC Sample Type	Rep 1		Rep 2		RPD (%)
% TOC	%	I-2 SED UP & LWR	Field Replicate	4.5		4.5		0%
Arsenic	mg/Kg dw	I-2 SED UP & LWR	Field Replicate	5.16		5.55		7%
Cadmium	mg/Kg dw	I-2 SED UP & LWR	Field Replicate	0.28		0.28		0%
Copper	mg/Kg dw	I-2 SED UP & LWR	Field Replicate	22.4		21.5		4%
Nickel	mg/Kg dw	I-2 SED UP & LWR	Field Replicate	21.9		22.4		2%
Lead	mg/Kg dw	I-2 SED UP & LWR	Field Replicate	20.1		21.1		5%
Zinc	mg/Kg dw	I-2 SED UP & LWR	Field Replicate	151		155		3%
Lube Oil	mg/Kg dw	I-2 SED UP & LWR	Field Replicate	870		760		13%
Benz[a]anthracene	ug/Kg dw	I-2 SED UP & LWR	Field Replicate	88	J	75	J	16%
Benzo(a)pyrene	ug/Kg dw	I-2 SED UP & LWR	Field Replicate	180	J	160	J	12%
Benzo(b)fluoranthene	ug/Kg dw	I-2 SED UP & LWR	Field Replicate	260		210	J	21%
Benzo(ghi)perylene	ug/Kg dw	I-2 SED UP & LWR	Field Replicate	360	J	310	J	15%
Benzo(k)fluoranthene	ug/Kg dw	I-2 SED UP & LWR	Field Replicate	130	J	120	J	8%
Chrysene	ug/Kg dw	I-2 SED UP & LWR	Field Replicate	190	J	160	J	17%
Fluoranthene	ug/Kg dw	I-2 SED UP & LWR	Field Replicate	240	J	220	J	9%
Indeno(1,2,3-cd)pyrene	ug/Kg dw	I-2 SED UP & LWR	Field Replicate	270	J	230	J	16%
Phenanthrene	ug/Kg dw	I-2 SED UP & LWR	Field Replicate	100	J	84	J	17%
Pyrene	ug/Kg dw	I-2 SED UP & LWR	Field Replicate	260		210	J	21%

UOM = Unit of measurement
QC = Quality control
RPD = Relative percent difference
Gray highlighted results represent RPDs above 20%.

Groundwater

MQOs for groundwater chemistry are shown in Table D-2. The BNA, PAH, and OPAH analyses had some results that were outside of QC limits. Results for these analyses were used as qualified by MEL.

An equipment blank was analyzed with the groundwater samples. It was processed by running deionized water from MEL through new Silastic tubing and a new QED QuickFilter® (model FF8100) at the cleaning room lab at Ecology Headquarters in Olympia. The equipment blank was relatively clean with no detections of metals, PAHs, and OPAHs (Appendix E, Table E-11). The BNA analysis (including TICs) for the equipment blank had a similar detection frequency to the samples at 16% (Table E-12).

Appendix E. Data Tables

Table E-1. Select diatom metrics, predicted direction, and upstream to downstream direction of metric response to increasing stress for Indian Creek sample sites.

Category		Metric	Definition	**Predicted response to increasing stress**	Metric response from I-1 to I-2
General indicators of overall stream health	General measure of water quality and habitat	Species Richness	Number of species in a sample	Decrease	Decrease
		Shannon Diversity Index	Function of both the number of species in a sample and the distribution of individuals	Decrease	Decrease
		Pollution Index	A weight average of species abundance and pollution tolerance rating within a sample	Increase	Decrease
		Percent Dominant Species	Percent of dominance of the single most abundant taxon	Increase	Increase
Stressor metrics	Sediment indicator taxa	Percent Motile Taxa	Taxa that are able to move from silt	Increase	Decrease*****
		Siltation Taxa Percent	Taxa that are tolerant of fine sediment	Increase	Decrease
	Metal Indicator Metrics	Heavy Metals Index	Percent abundance of metals-tolerant taxa	Increase	Increase*
		Percent *Eunotia* individuals	Percent of diatom species from the genus *Eunotia*	Increase	Increase*
		Percent acidophilus	Occur in water with a pH < 7	Increase	Increase*

*Indicates differences were greater than coefficients of variation (CVs) from pooled duplicate samples.

Table E-2. Select macroinvertebrate metrics, predicted direction, and upstream to downstream direction of metric response to increasing stress for Indian Creek sample sites.

Category		Metric	Definition	**Predicted response to increasing stress**	Metric response from I-1 to I-2
General indicators of overall stream health	**Richness Measures:** General measure of water quality and habitat	Total No. taxa	Number of species in a sample	Decrease	Increase
		No. Ephemeroptera taxa	Number of mayfly taxa	Decrease	Increase*
		No. Plecoptera taxa	Number of stonefly taxa	Decrease	Increase
		No. Trichoptera taxa	Number of caddisfly taxa	Decrease	Increase*
		% EPT	Percent of the composite of mayfly, stonefly, and caddisfly larvae	Decrease	Decrease*
	Tolerance/Intolerance **Measures:** General measure of stress	No. Intolerant taxa	Richness of taxa sensitive to perturbation	Decrease	Decrease
		% Supertolerant	Percent of taxa tolerant of perturbation	Increase	Increase*
		% Dominant taxon	Measures the dominance of the single most abundant taxon	Increase	Increase*
		Hilsenhoff Biotic Index	Uses tolerance values to weight abundance in an estimate of overall pollution	Increase	Increase
	Life cycle measures: Generally non-specific measure of stress	% Multivoltine	Percent of organisms having short life cycle	Increase	Increase
		% Univoltine	Percent of organisms relatively long-lived	Decrease	Increase*
Stressor Metrics	**Feeding measures:** Indicators of trophic conditions	% Grazers and Scrapers	Percent of taxa that scrape or graze upon periphyton	Decrease	Decrease
	Habit measures: Indicators of sediment	No. Clinger Taxa	Number of clinger taxa	Decrease	Increase
		% Clingers	Percent of insects having fixed retreats	Decrease	Decrease
	Metal Tolerance: Indicators of metals	% Metal tolerant taxa	Percent abundance of metals-tolerant taxa	Increase	Increase

*Indicates differences were greater than coefficients of variation (CVs) from pooled duplicate samples.

Table E-3. Select diatom metrics, upstream to downstream direction of metric response to increasing stress for Indian Creek sample sites, coefficients of variation (CVs) between sample sites, and metric CVs from replicate sites.

Metric	Upstream	Downstream	Response	CV	Replicate CV
Stream Health Indicator Metrics					
Species Richness	41	41	None	-	7.4
Shannon Diversity Index	3.81	3.70	Decrease	2.11	4.3
Pollution Index	2.32	2.25	Decrease	1.9	10.3
Percent Dominant Species	23.8	25.4	Increase	4.7	30.2
Sediment Indicator Metrics					
Percent Siltation Taxa	36.8	23.2	Decrease	32.0	40.7
Percent Motile Taxa	40.8	25.3	Decrease	33.0	14.2
Metals Indicator Metrics					
Percent Metal Tolerant Taxa	27.0	34.5	Increase	17.2	15.6
Percent *Eunotia*	1.4	2.2	Increase	29.7	25.6
Percent Acidophilus	0.3	1.4	Increase	80.5	75.0

Table E-4. Select macroinvertebrate metrics collected with D-net, upstream to downstream direction of metric response to increasing stress for Indian Creek sample sites, coefficients of variations (CVs) between sample sites and metric CVs from replicate sites.

Metric	Upstream	Downstream	Response	CV	Replicate CV
Stream Health Indicator Metrics					
BIBI	25.7	26.7	Increase	2.7	8.10
Total Taxa Richness	36	30	Decrease	11.8	16.8
Ephemeroptera Richness	3.1	3.5	Increase	17.9	1.2
Plecoptera Richness	2.3	3.6	Increase	9.4	53.9
Trichoptera Richness	4.2	5.3	Increase	17.4	12.6
% EPT	74	57	Decrease	17.9	10.8
% Intolerant taxa	34.5	25.7	Decrease	20.5	20.5
Percent tolerant organisms	2.1	2.1	None	3.65	77.8
Percent supertolerant	3.6	11.0	Increase	72.4	31.9
Percent dominant taxon	25.9	32.6	Increase	24.0	16.5
Hilsenhoff Biotic Index	3.2	3.9	Increase	1.5	13.9
Percent Multivoltine	22.5	35.6	Increase	24.6	31.6
Percent Univoltine	14.3	18.7	Increase	23.8	18.6
Sediment Indicator Metrics					
Clinger Richness	10.8	12.5	Increase	8.8	10.1
Percent Clingers	62.3	47.4	Decrease	9.0	19.1
Metals Indicator Metrics					
Metal Tolerance Index	3.7	2.7	Decrease	2.8	6.8

Table E-5. 2013 metric values using D-net and bug bags in lower Indian Creek*.

Metric Values	D-net	Bug bags
Total Taxa Richness	40	48
Ephemeroptera Richness	5	4
Plecoptera Richness	2	4
Trichoptera Richness	5	2
Pollution Sensitive Richness	1	2
Clinger Richness	10	11
Semivoltine Richness	3	4
Pollution Tolerant Percent	0.20%	2.60%
Predator Percent	6.47%	4.65%
Dominant Taxa (3) Percent	68.82%	56.32%
BIBI	24	26

*The location ID in EIM is called IND-2.5 for invertebrates collected by D-net.
The EIM location ID for bug bags is called I-2C.

Table E-6. Measurements for Surface Water, Groundwater, and Stormwater collected in spring of 2013.

Matrix	EIM Location ID	Date	Time	Temp. (deg C)	Conductivity (umhos/cm)	pH	DO (%Sat)	DO (mg/L)	Flow (CFS)	Co-occurring activity
Surface Water	INDIAN-1	4/24/13	10:15	9.03	115.1	6.84	97.8	11.44	1.81	Macroinvertebrate and periphyton collection
	"	4/30/13	12:20	10	121.4	7.04	98.5	11.3	base	Trout basket placement
	"	5/7/13	11:40	12.45	131.6	7.16	96.4	10.41	base	Trout water level check
	"	5/17/13	12:45	12.51	134.0	7.25	NA	10.46	NA	Trout Check and replicate 1 taken for metals analysis (alevin stage)
	"	5/30/13	11:00	11.46	124.8	6.99	NA	10.64	NA	Trout test termination (fry stage)
	INDIAN-2B	4/23/13	10:55	9.2	134.7	7.16	NA	NA	base	Collection of surface water sample at 2010 passive sampler location
	INDIAN-2C	4/24/13	13:20	10.56	124.5	6.9	97.1	10.95	3.23	Macroinvertebrate and periphyton collection
	"	4/25/13	13:35	11.24	133.1	6.78	95.7	10.65	NA	Surface water adjacent to seep sampling location (Indian GW-1)
	INDIAN-2B	4/26/13	14:05	11.90	135.3	7.08	95.7	10.49	NA	Surface water adjacent to suspect culvert (QUINCE STW) at stream baseflow
	INDIAN-2C	4/26/13	15:15	12.35	134.4	7.21	96.0	10.39	NA	Surface water adjacent to seep sampling location (Indian GW-2)
	"	5/2/13	12:15	10.33	140.1	7.04	97.1	11.0	NA	Bug bag placement
	INDIAN-2B	5/13/13	15:05	13.78	136.0	7.17	NA	9.85	NA	Surface water adjacent to suspect culvert (QUINCE STW) at latter part of storm event
	"	5/13/13	18:43	13.77	131.4	7.15	92.1	9.61	NA	Surface water adjacent to suspect culvert (QUINCE STW) post storm event
	"	6/12/13	15:22	13.89	140.1	6.92	93.5	9.61	NA	Surface water sample at 2010 passive sampler location (post storm)
	"	6/12/13	17:40	12.63	144.3	7.13	95.8	10.12	NA	Surface water measurement at 2010 passive sampler location (post storm)
	INDIAN-2A	4/30/13	13:10	10.05	136.9	6.85	94.6	10.8	base	Trout basket placement
	"	5/7/13	10:55	12.3	148.4	7.11	92.0	9.92	base	Trout water level check
	"	5/17/13	12:00	12.35	150.9	7.18	NA	9.93	NA	Trout Check and replicate 1 taken for metals analysis (alevin stage)
	"	5/30/13	12:20	11.55	135.3	7.11	NA	10.17	NA	Trout test termination (fry stage)

Matrix	EIM Location ID	Date	Time	Temp. (deg C)	Conductivity (umhos/cm)	pH	DO (%Sat)	DO (mg/L)	Flow (CFS)	Co-occurring activity
	INDIAN-2B	4/30/13	13:35	10.11	137.7	7.14	95.6	10.9	base	Trout basket placement
	"	5/7/13	10:45	12.34	149.8	7.12	94.1	10.11	base	Trout water level check
	"	5/17/13	11:40	12.38	151.6	7.21	NA	10.09	NA	Trout Check and test termination (alevin stage)
	INDIAN-2C	4/30/13	14:10	10.29	137.9	7.23	96.6	11.01	base	Trout basket placement
	"	5/7/13	11:00	12.33	149.9	7.17	93.9	10.16	base	Trout water level check
	"	5/17/13	10:55	12.29	152.3	7.18	NA	10.17	NA	Trout Check and test termination (alevin stage)
Ground-water	INDIAN GW-1	4/25/13	14:45	13.32	260.6	6.85	27.3	2.88	base	Groundwater measurement prior to sample taken at 1500
	"	"	14:48	13.03	260.7	6.82	26.8	2.83	"	"
	"	"	14:51	13.33	260.9	6.83	22.4	2.37	"	"
	"	"	15:47	13.34	258.7	6.92	22.4	2.08	"	Groundwater measurement after sample taken at 1500
	"	"	15:50	12.55	259.3	6.8	23.4	2.56	"	"
	"	"	15:56	12.51	260.3	6.71	17.7	1.90	"	"
	QUINCE STW	4/26/13	14:12	14.22	236.4	7.01	NA	NA	base	Measurement of baseflow from suspect culvert - sample taken at 1425
	"	"	14:15	14.23	235.8	7.13	74.8	7.76	"	"
	"	"	14:18	14.23	236.1	7.04	74.1	7.68	"	"
	INDIAN GW-2	4/26/13	15:19	11.69	241.7	7.35	17.4	1.90	base	Groundwater measurement prior to sample taken at 1535
	"	"	15:22	11.68	242.2	7.19	16.7	1.83	"	"
	"	"	15:25	11.62	241.7	7.32	16.6	1.82	"	"
	"	"	15:56	11.76	241.9	7.50	15.4	1.69	"	Groundwater measurement after sample taken at 1535
	"	"	15:59	11.72	241.5	7.48	18.0	1.97	"	"

Matrix	EIM Location ID	Date	Time	Temp. (deg C)	Conductivity (umhos/cm)	pH	DO (%Sat)	DO (mg/L)	Flow (CFS)	Co-occurring activity
Storm-water	QUINCE STW	5/13/13	15:15	18.16	129.6	6.72	64.5	6.15	storm	Stormwater from suspect culvert (QUINCE STW)
	"	"	18:43	16.88	182.7	6.83	57.1	5.55	base	Water from suspect culvert (QUINCE STW) post storm baseflow
	"	6/12/13	15:00	16.75*	86.5*	6.64*	78*	7.55*	storm	Stormwater from suspect culvert (QUINCE STW)
	PLUM STW	"	15:08	16*	66.5*	6.76*	84.6*	8.3*	storm	Stormwater from downstream stormwater culvert (PLUM STW)

* Estimated results as measurements were taken from a bottle 30 minutes after sample collection.
EIM = Environmental Information Management database.
DO = Dissolved oxygen
NA = Not analyzed

Table E-7. Chemical Results for Lower Indian Creek Sediment Traps.

Sample ID	I-2 SED UP		I-2 SED LWR		I-2 SED REP	
Sample No.	1306040-01		1306040-02		1306040-03	
Deployment	5/2/2013 - 6/20/13					
% Solids	**48.7**		**54.2**		**53.9**	
TOC %	**4.5**		**4.5**		**3.9**	
Metals (mg/Kg dw)						
Arsenic	**5.16**		**5.55**		**4.84**	
Cadmium	**0.28**		**0.28**		**0.28**	
Copper	**22.4**		**21.5**		**20.1**	
Nickel	**21.9**		**22.4**		**22.1**	
Lead	**20.1**		**21.1**		**19.9**	
Zinc	**151**		**155**		**147**	
Total Petroleum Hydrocarbons (mg/Kg dw)						
#2 Diesel	29	U	27	U	28	U
Lube Oil	**870**		**760**		**920**	
PAHs (ug/Kg dw)						
1-Methylnaphthalene	500	U	460	U	460	U
2-Chloronaphthalene	500	U	460	U	460	U
2-Methylnaphthalene	500	U	460	U	460	U
Acenaphthene	250	U	230	U	230	U
Acenaphthylene	250	U	230	U	230	U
Anthracene	250	U	230	U	230	U
Benz[a]anthracene	**88**	**J**	**75**	**J**	**140**	**J**
Benzo(a)pyrene	**180**	**J**	**160**	**J**	**200**	**J**
Benzo(b)fluoranthene	**260**		**210**	**J**	**280**	
Benzo(ghi)perylene	**360**	**J**	**310**	**J**	**310**	**J**
Benzo(k)fluoranthene	**130**	**J**	**120**	**J**	**160**	**J**
Carbazole		REJ		REJ		REJ
Chrysene	**190**	**J**	**160**	**J**	**220**	**J**
Dibenzo(a,h)anthracene	500	U	460	U	460	U
Dibenzofuran	500	U	460	U	460	U
Fluoranthene	**240**	**J**	**220**	**J**	**340**	
Fluorene	250	U	230	U	230	U
Indeno(1,2,3-cd)pyrene	**270**	**J**	**230**	**J**	**250**	**J**
Naphthalene	500	U	460	U	460	U
Phenanthrene	**100**	**J**	**84**	**J**	**140**	**J**
Pyrene	**260**		**210**	**J**	**330**	
Retene	250	U	430		230	U
Non-Detected BNAs (ug/Kg dw)						
1,2,4-Trichlorobenzene	500	U	460	U	460	U
1,2-Dichlorobenzene	1000	U	920	U	920	U

Sample ID	I-2 SED UP		I-2 SED LWR		I-2 SED REP	
Sample No.	1306040-01		1306040-02		1306040-03	
Deployment	5/2/2013 - 6/20/13					
1,2-Diphenylhydrazine	250	U	230	U	230	U
1,3-Dichlorobenzene	1000	U	920	U	920	U
1,4-Dichlorobenzene	1000	U	920	U	920	U
2,4,5-Trichlorophenol	1000	U	920	U	920	U
2,4,6-Trichlorophenol	1000	U	920	U	920	U
2,4-Dichlorophenol	2500	U	2300	U	2300	U
2,4-Dimethylphenol	2500	U	2300	U	2300	U
2,4-Dinitrophenol		REJ		REJ		REJ
2,4-Dinitrotoluene	1000	U	920	U	920	U
2,6-Dinitrotoluene	1000	U	920	U	920	U
2-Chlorophenol	1000	U	920	U	920	U
2-Methylphenol	2500	U	2300	U	2300	U
2-Nitroaniline		REJ	4600	U	4600	U
2-Nitrophenol	500	UJ	460	UJ	460	UJ
3,3'-Dichlorobenzidine		REJ	920	UJ	920	UJ
3B-Coprostanol	5000	UJ	4600	UJ	4600	UJ
3-Nitroaniline		REJ		REJ		REJ
4,6-Dinitro-2-Methylphenol	1000	UJ	920	UJ	920	UJ
4-Bromophenyl phenyl ether	500	U	460	U	460	U
4-Chloro-3-Methylphenol	2500	U	2300	U	2300	U
4-Chloroaniline		REJ		REJ		REJ
4-Chlorophenyl-Phenylether	250	U	230	U	230	U
4-Methylphenol	2500	U	2300	U	2300	U
4-Nitroaniline		REJ	920	UJ	920	UJ
4-Nitrophenol	2500	U	2300	U	2300	U
4-nonylphenol	1000	U	920	U	920	U
Benzoic Acid		REJ		REJ		REJ
Benzyl Alcohol		REJ	2300	UJ	2300	UJ
Bis(2-chloro-1-methylethyl) ether	250	U	230	U	230	U
Bis(2-Chloroethoxy)Methane	250	U	230	U	230	U
Bis(2-Chloroethyl)Ether	500	U	460	U	460	U
Bis(2-Ethylhexyl) Phthalate	2400	UJ	1700	U	1700	U
Bisphenol A	2500	UJ	230	UJ	230	UJ
Butyl benzyl phthalate	500	U	460	U	460	U
Caffeine	500	UJ	460	UJ	460	UJ
Cholesterol	5000	UJ	4600	UJ	4600	UJ
Diethyl phthalate	250	U	230	U	230	U
Dimethyl phthalate	250	U	230	U	230	U
Di-N-Butylphthalate	250	U	230	U	230	U
Di-N-Octyl Phthalate	2500	U	2300	U	2300	U

Sample ID	I-2 SED UP		I-2 SED LWR		I-2 SED REP	
Sample No.	1306040-01		1306040-02		1306040-03	
Deployment	5/2/2013 - 6/20/13					
Hexachlorobenzene	250	U	230	U	230	U
Hexachlorobutadiene	1000	U	920	U	920	U
Hexachlorocyclopentadiene	1000	UJ	920	UJ	920	UJ
Hexachloroethane	250	U	230	U	230	U
Isophorone	500	U	460	U	460	U
Nitrobenzene	250	U	230	U	230	U
N-Nitrosodi-n-propylamine	250	U	230	U	230	U
N-Nitrosodiphenylamine	500	UJ	460	UJ	460	UJ
Pentachlorophenol	500	U	460	U	460	U
Phenol	1000	U	920	U	920	U
Triclosan	1000	UJ	920	UJ	920	UJ
Triethyl citrate	250	UJ		REJ		REJ

Site Descriptions:
I-2 SED UP: 14 ft downstream of trout location I-2C
I-2 SED LWR: 21 ft downstream of I-2 SED UP
I-2 SED REP: Split processing sample of I-2 SED LWR

Bolded values indicate detected results
J = Analyte was positively identified; reported result is an approximate concentration
U = Not detected above the reported quantitation limit
UJ = Not detected above the reported estimated quantitation limit
REJ = Result rejected due to quality control failures

Table E-8. Tentatively Identified Chemical Compounds from the BNA Analysis of Sediment Trap Samples from Lower Indian Creek, ug/Kg dw.

Sample ID	I-2 SED UP		I-2 SED LWR		I-2 SED REP	
Sample No.	1306040-01		1306040-02		1306040-03	
Deployment	5/2/2013 - 6/20/13					
1-Hentetracontanol	ND		**2000**	**NJ**	ND	
1-Iodo-2-methylundecane	ND		ND		**210**	**NJ**
1-Nonadecene	**820**	**NJ**	ND		ND	
1-Octadecene	**880**	**NJ**	**1300**	**NJ**	ND	
1-Octadecene(1)	**1500**	**NJ**	ND		ND	
13-Octadecenal	**12000**	**NJ**	ND		ND	
14-Isocopalene	**1000**	**NJ**	ND		ND	
14-Octadecenal	**7500**	**NJ**	ND		ND	
17-Octadecenal	ND		**900**	**NJ**	ND	
17-Octadecenal(1)	ND		**5000**	**NJ**	ND	
17-Pentatriacontene	ND		ND		**5500**	**NJ**
28-Nor-17.alpha.(H)-hopane	**1600**	**NJ**	**2400**	**NJ**		
5-Octadecene, (E)-	ND		ND		**130**	**NJ**
8-(1,1,2-Trimethyl-2-propenyl)-8H-cycloh	ND		**6800**	**NJ**	ND	
Acetic acid, octadecyl ester	ND		**1900**	**NJ**	ND	
Anthracene, 9-dodecyltetradecahydro-	ND		**2900**	**NJ**	ND	
Bicyclo[3.1.0]hexane, 4-methylene-1-(1-m	ND		ND		**430**	**NJ**
Cyclopropane, 1-methyl-1-(1-methylethyl)	**1700**	**NJ**	ND		ND	
Cyclopentanecarboxylic acid, 2-amino-, c	ND		ND		**180**	**NJ**
Cyclopentene, 1,3-dimethyl-2-(1-methylet	ND		ND		**110**	**NJ**
Cyclotetracosane	**6000**	**NJ**	ND		**4300**	**NJ**
Eicosane, 10-methyl-	**730**	**NJ**	ND		ND	
Eicosane	ND		ND		**310**	**NJ**
Heneicosane	ND		**1200**	**NJ**	**1700**	**NJ**
Heptacosane	ND		**6300**	**NJ**	**700**	**NJ**
Hexadecane	**2000**	**NJ**	**810**	**NJ**	**16000**	**NJ**
Hexadecane(1)	**12000**	**NJ**	ND		ND	
Hexadecanoic acid	**1800**	**NJ**	ND		**530**	**NJ**
Hexatriacontane	**13000**	**NJ**	ND		**17000**	**NJ**
Naphthalene, 1,2,3,5,6,7,8,8a-octahydro-	**1600**	**NJ**	**3100**	**NJ**	ND	
Nonacosane	ND		**5000**	**NJ**	ND	
Nonadecane	ND		**660**	**NJ**	ND	
Nonanal	ND		ND		**160**	**NJ**
NOROLEAN-12-ENE	ND		**14000**	**NJ**	ND	
Octacosane	ND		**880**	**NJ**	ND	
Octadecane	**1100**	**NJ**	**900**	**NJ**	**220**	**NJ**
Olean-12-Ene	**16000**	**NJ**	ND		**13000**	**NJ**
Olean-13(18)-ene	**24000**	**NJ**	ND		ND	

Sample ID	I-2 SED UP		I-2 SED LWR		I-2 SED REP	
Sample No.	1306040-01		1306040-02		1306040-03	
Deployment	5/2/2013 - 6/20/13					
Pentacosane	**4700**	**NJ**	**2900**	**NJ**	**3400**	**NJ**
Pentadecanal-	**1300**	**NJ**	**1100**	**NJ**	**860**	**NJ**
Phenanthrene, 1,2,3,4,4a,9,10,10a-octahy	**3100**	**NJ**	ND		ND	
Stigmast-4-en-3-one	**2500**	**NJ**	**2500**	**NJ**	ND	
Tetracosane	**1300**	**NJ**	**1100**	**NJ**	ND	
(Z)14-Tricosenyl formate	ND		**8900**	**NJ**	ND	
Z-3-(1-Methylpropyliden)bicyclo[2.2.1]he	ND		ND		**140**	**NJ**
Unknown Hydrocarbon 1 20.225	ND		ND		**190**	**NJ**
Unknown Hydrocarbon 3 31.293	ND		ND		**15000**	**NJ**
Unknown Hydrocarbon 32.733	ND		**12000**	**NJ**	ND	

Site Descriptions:
I-2 SED UP: 14 ft downstream of trout location I-2C
I-2 SED LWR: 21 ft downstream of I-2 SED UP
I-2 SED REP: Split processing sample of I-2 SED LWR

Bold values indicate detected results
ND = Not detected (qualitative)
NJ = Parameter is tentatively identified and the associated results value is an estimate
BNA = Bases, neutrals, and acids

Table E-9. Chemical Results for Surface Water from Lower Indian Creek.

Sample ID	I-2 SW		I-2 SW dup		I-2 SW	
Sample No.	1304070-01		1304070-02		1306055-03	
Date	4/23/2013		4/23/2013		6/12/2013	
Time	1015		1055		1522	
Total Suspended Solids (mg/L)	**6**		**6**		**208**	
DOC (mg/L)	**6.5**		**6.5**		**14.6**	
Alkalinity (mg/L)	**49.1**		**49.3**		**51.0**	
Hardness (mg/L)	**52.5**		**52.5**		**58.0**	
Metals (ug/L)						
Arsenic - total	**0.74**		**0.73**		**3.55**	
Arsenic - dissolved	**0.62**		**0.62**		**0.57**	
Cadmium - total	0.10	U	0.10	U	**0.28**	
Cadmium - dissolved	0.02	U	0.02	U	**0.025**	
Copper - total	**1.12**		**1.16**		**33.1**	
Copper - dissolved	**0.65**		**0.66**		**7.96**	
Nickel - total	**1.32**		**1.32**		**8.98**	
Nickel - dissolved	**1.09**		**1.10**		**1.56**	
Lead - total	**0.52**		**0.53**		**12.4**	
Lead - dissolved	**0.178**		**0.172**		**0.242**	
Zinc -total	**5.2**		**5.5**		**192**	
Zinc - dissolved	**2.9**		**2.9**		**12.2**	
PAHs (ug/L)						
1-Methylnaphthalene	0.010	U	0.010	U	0.010	U
2-Chloronaphthalene	0.010	U	0.010	U	0.010	U
2-Methylnaphthalene	0.010	U	0.010	U	0.010	U
Acenaphthene	**0.0075**	**J**	0.010	U	**0.0069**	**J**
Acenaphthylene	0.010	U	0.010	U	**0.033**	
Anthracene	0.010	U	0.010	U	**0.011**	**NJ**
Benz[a]anthracene	0.010	U	0.010	U	0.024	U
Benzo(a)pyrene	0.010	U	0.010	U	**0.031**	**J**
Benzo(b)fluoranthene	0.010	U	0.010	U	**0.032**	
Benzo(ghi)perylene	0.010	U	0.010	U	**0.039**	
Benzo(k)fluoranthene	0.010	U	0.010	U	**0.024**	
Carbazole	0.010	UJ	0.010	UJ	0.010	U
Chrysene	0.010	U	0.010	U	**0.039**	
Dibenzo(a,h)anthracene	0.010	U	0.010	U	0.015	UJ
Dibenzofuran	0.010	UJ	0.010	UJ	0.010	U
Fluoranthene	0.010	U	0.010	U	**0.044**	
Fluorene	0.010	UJ	0.010	UJ	0.010	UJ
Indeno(1,2,3-cd)pyrene	0.010	U	0.010	U	0.032	UJ
Naphthalene	**0.015**		**0.013**		0.010	U
Phenanthrene	0.010	U	0.010	U	**0.019**	

Sample ID	I-2 SW		I-2 SW dup		I-2 SW	
Sample No.	1304070-01		1304070-02		1306055-03	
Date	4/23/2013		4/23/2013		6/12/2013	
Time	1015		1055		1522	
Pyrene	0.010	U	0.010	U	**0.058**	**J**
Retene	0.010	U	0.010	U	**0.015**	**J**
Oxygenated PAHs (ug/L)						
1,4-Anthraquinone	REJ		REJ		REJ	
4H-Cyclopenta[def]phenanthren-4-one	0.021	U	0.021	U	**0.013**	**J**
5,12-Naphthacenequinone	0.021	UJ	0.021	UJ	0.020	UJ
7,12-Benz[a]anthracenquinone	0.021	UJ	0.021	UJ	0.020	UJ
9,10-Anthracenedione	0.051	U	0.051	U	0.050	UJ
9,10-Phenthrenequinone	REJ		REJ		0.020	U
9H-Fluoren-9-one	0.051	U	0.051	U	0.050	UJ
Aceanthracenequinone	0.051	U	0.051	U	0.050	U
Acenaphthenequinone	0.10	UJ	0.10	UJ	0.099	U
Benzanthrone	0.021	U	0.021	U	0.020	UJ
Benzo[a]fluorenone	0.021	U	0.021	U	**0.029**	
Benzo[c]phenanthrene-1[1,4]quinone	REJ		REJ		REJ	
Benzo[cd]pyrenone	0.021	UJ	0.021	UJ	0.020	UJ
Phenanthrene-1,4-dione	REJ		REJ		REJ	
Pesticides (ug/L)						
Captan	0.034	U	0.035	U	0.034	U
Tetrahydrophthalimide (THPI)	0.10	U	0.11	U	0.10	U
Tentatively Identified Compounds (qualitative)						
benzenepropanioc acid, 3,5-bis(1,1-dimethylethyl)-4-hydroxy methyl ester	ND		**Detect**		ND	
Benzoic acid, 3-amino-, methyl ester					**Detect**	
Dichlobenil	**Detect**		**Detect**		ND	
Tebuthiuron	ND		**Detect**		ND	
1-phenanthrenecarboxylic acid	ND		ND		**Detect**	
1-undecene, 7-methyl-	ND		**Detect**		ND	
2,3,4-trimethyl hexane	**Detect**		ND		ND	
2,5-cyclohexadiene-1 one,2,6-bis(1,1-dimethylethyl)-4-ethylidene	**Detect**		ND			
2,6-dichloro benzimide	ND		**Detect**		**Detect**	
3,5-dichloro-N-(1,1-dimethylprolynyl) benzamide	**Detect**		**Detect**		ND	
phthalic acid, 6-ethyl-3-octyl isobutyl ester	**Detect**		**Detect**		ND	
phthalic acid, isobutyl tridecyl ester	ND		ND		**Detect**	
Total Petroleum Hydrocarbons	**Detect**		**Detect**		**Detect**	

Notes for Table E-9

Site Descriptions and Comments:
I-2 SW: Same location as the 2010 organics passive samplers (nearest location I-2B in 2013)
4/23/2013 – Baseflow conditions in the creek
6/12/2013 – Storm runoff event affecting the creek

Bolded values indicate detected results
J = Analyte was positively identified; reported result is an approximate concentration
U = Not detected above the reported quantitation limit
UJ = Not detected above the reported estimated quantitation limit
NJ = Analyte was tentatively identified; reported result is an approximate concentration
ND = Not detected (qualitative)
NA = Not analyzed
REJ = Result rejected due to quality control failures

Table E-10. Chemical Results for Stormwater near Lower Indian Creek.

Location	QUINCE STW		QUINCE STW		PLUM STW		Trip Blank	
Sample No.	1305036-01		1306055-01		1306055-02		1306055-04	
Date	5/13/2013		6/12/2013		6/12/2013		6/12/2013	
Time	1515		1500		1508		1755	
Total Suspended Solids (mg/L)	**3**		**25**		**39**		NA	
TOC (mg/L)	**8.0**		NA		NA		NA	
DOC (mg/L)	**5.9***		**18.8**		**32.7**		NA	
Alkalinity (mg/L)	**50.3**		**30.9**		**5.9**		NA	
Hardness (mg/L)	**49.7**		**35.7**		**15.6**		0.30	U
Metals (ug/L)								
Arsenic - total	**0.82**		**1.26**		**0.98**		0.10	U
Arsenic - dissolved	**0.86**		**0.71**		**0.69**		0.10	U
Cadmium - total	0.10	U	0.10	U	**0.16**		0.10	U
Cadmium - dissolved	0.02	U	**0.025**		**0.086**		0.02	U
Copper - total	**3.61**		**10.0**		**58.2**		0.10	U
Copper - dissolved	**3.45**		**6.92**		**36.3**		0.10	U
Nickel - total	**0.76**		**2.47**		**4.99**		0.10	U
Nickel - dissolved	**0.69**		**1.77**		**2.72**		0.10	U
Lead - total	**0.21**		**2.17**		**5.03**		0.10	U
Lead - dissolved	**0.187**		**0.203**		**0.386**		0.02	U
Zinc -total	**9.5**		**39.0**		**198**		5.0	U
Zinc - dissolved	**9.0**		**29.5**		**124**		1.0	U
PAHs (ug/L)								
1-Methylnaphthalene	**0.019**		**0.003**	**NJ**	**0.010**	**NJ**	0.011	U
2-Chloronaphthalene	0.010	U	0.010	U	0.010	U	0.011	U
2-Methylnaphthalene	**0.014**		**0.010**	**NJ**	0.010	U	0.011	U
Acenaphthene	**0.097**		**0.031**		0.010	U	0.011	U
Acenaphthylene	0.010	U	0.010	U	**0.085**	**NJ**	0.011	U
Anthracene	**0.015**		**0.018**	**NJ**	**0.017**	**NJ**	0.011	U
Benz[a]anthracene	0.010	U	**0.054**	**J**	0.023	U	0.011	U
Benzo(a)pyrene	**0.015**		**0.087**	**J**	**0.039**	**J**	0.011	U
Benzo(b)fluoranthene	**0.011**		**0.12**		**0.058**		0.011	U
Benzo(ghi)perylene	**0.013**		**0.088**		**0.086**		0.011	U
Benzo(k)fluoranthene	**0.0075**	**J**	**0.080**		**0.032**		0.011	U
Carbazole	0.047	U	0.054	UJ	0.010	U	0.011	U
Chrysene	**0.014**		**0.11**		**0.094**		0.011	U
Dibenzo(a,h)anthracene	0.010	U	0.027	UJ	0.018	UJ	0.011	U
Dibenzofuran	**0.053**	**NJ**	**0.020**	**NJ**	0.010	U	0.011	U
Fluoranthene	**0.038**		**0.13**		**0.064**		0.011	U
Fluorene	**0.046**	**J**	**0.018**	**NJ**	0.010	UJ	0.011	UJ
Indeno(1,2,3-cd)pyrene	**0.015**		**0.085**	**J**	0.042	UJ	0.011	U
Naphthalene	**0.044**		0.015	U	0.017	U	0.011	U

Location	QUINCE STW	QUINCE STW	PLUM STW	Trip Blank
Sample No.	1305036-01	1306055-01	1306055-02	1306055-04
Date	5/13/2013	6/12/2013	6/12/2013	6/12/2013
Time	1515	1500	1508	1755
Phenanthrene	**0.028**	**0.045**	**0.039**	0.011 U
Pyrene	**0.027**	**0.12**	**0.13**	0.011 U
Retene	0.010 U	0.010 U	**0.034 J**	0.011 U
Oxygenated PAHs (ug/L)				
1,4-Anthraquinone	REJ	REJ	REJ	REJ
4H-Cyclopenta[def]phenanthren-4-one	**0.017 J**	**0.017 J**	0.020 UJ	0.021 UJ
5,12-Naphthacenequinone	0.020 UJ	**0.076 J**	0.020 UJ	0.021 UJ
7,12-Benz[a]anthracenquinone	0.020 U	**0.069 J**	0.020 UJ	0.021 UJ
9,10-Anthracenedione	0.050 UJ	0.050 UJ	0.065 UJ	0.053 UJ
9,10-Phenthrenequinone	REJ	REJ	REJ	REJ
9H-Fluoren-9-one	**0.062 J**	**0.064 J**	0.050 UJ	0.053 UJ
Aceanthracenequinone	0.050 U	0.050 UJ	0.050 UJ	0.053 UJ
Acenaphthenequinone	0.10 U	0.099 U	0.10 U	0.11 U
Benzanthrone	**0.018 J**	0.020 UJ	0.020 UJ	0.021 UJ
Benzo[a]fluorenone	**0.020 J**	**0.055**	**0.069**	0.021 U
Benzo[c]phenanthrene-1[1,4]quinone	REJ	REJ	REJ	REJ
Benzo[cd]pyrenone	REJ	**0.057**	**0.048 J**	0.021 UJ
Phenanthrene-1,4-dione	REJ	REJ	REJ	REJ
Pesticides (ug/L)				
Captan	0.034 UJ	0.034 U	0.034 U	0.035 U
Tetrahydrophthalimide (THPI)	0.10 U	0.10 U	0.10 U	0.11 U
Tentatively Identified Compounds (qualitative)				
Dichlobenil	**Detect**	ND	ND	ND
Tebuthiuron	**Detect**	ND	ND	ND
Caffeine	**Detect**	ND	ND	ND
Fyrol PCF (1st peak)	**Detect**	ND	ND	ND
1H-inden-1-one, 2,3-dihydro-5,6-dimethoxy-3-methyl-	**Detect**	ND	ND	ND
1-phenanthrenecarboxylic acid	ND	**Detect**	**Detect**	ND
1,2-benzenedicarboxylic acid, butyl decyl ester	ND	ND	ND	**Detect**
2(3H)-benzothiazole	ND	ND	**Detect**	ND
Phthalimide	ND	ND	**Detect**	ND
Phenol, 2-(2H-benzotriazol-2-yl)-4,6-bis(1,1-dimethypropyl)-	ND	**Detect**	ND	ND
Thieno[2,30c]pyridine	ND	ND	**Detect**	ND
Total Petroleum Hydrocarbons	**Detect**	**Detect**	**Detect**	ND

Notes for Table E-10

Site Descriptions and Comments:
QUINCE STW: Stormwater collected from suspect culvert
PLUM STW: Stormwater culvert downstream of study area
Trip Blank: Laboratory deionized water transferred to bottles in the field
5/13/2013 – Stormwater sample taken at the tail end of a storm
6/12/2013 – Stormwater sample taken during the middle of a storm

* = DOC sample taken over an hour after (1620 hrs) the other storm samples were collected
Bolded values indicate detected results
J = Analyte was positively identified; reported result is an approximate concentration
U = Not detected above the reported quantitation limit
UJ = Not detected above the reported estimated quantitation limit
NJ = Analyte was tentatively identified; reported result is an approximate concentration
ND = Not detected (qualitative)
NA = Not analyzed
REJ = Result rejected due to quality control failures

Table E-11. Chemical Results for Groundwater in Lower Indian Creek.

Location	INDIAN GW-1		QUINCE STW		INDIAN GW-2		Equip. Blank	
Sample No.	1304069-01		1304069-02		1304069-03		1304069-04	
Date	4/25/2013		4/26/2013		4/26/2013		4/26/2013	
Time	1500		1425		1535		1700	
Alkalinity (mg/L)	**116**		**102**		**103**		NA	
DOC (mg/L)	**1.4**		**1.5**		1.0	U	NA	
Hardness (mg/L)	**115**		**101**		**108**		NA	
Dissolved Metals (ug/L)								
Arsenic	**4.84**		**1.03**		**2.53**		0.10	U
Cadmium	0.02	U	0.02	U	0.02	U	0.02	U
Copper	0.10	U	**0.21**		0.10	U	0.10	U
Nickel	**0.58**		**0.61**		**0.45**		0.10	U
Lead	0.02	U	0.02	U	0.02	U	0.02	U
Zinc	**1.1**		**2.1**		1.0	U	1.0	U
PAHs (ug/L)								
1-Methylnaphthalene	0.010	U	**0.030**		0.011	U	0.011	U
2-Chloronaphthalene	0.010	U	0.011	U	0.011	U	0.011	U
2-Methylnaphthalene	0.010	U	**0.018**		0.011	U	0.011	U
Acenaphthene	0.010	U	**0.13**		0.011	U	0.011	U
Acenaphthylene	0.010	U	0.011	U	0.011	U	0.011	U
Anthracene	0.010	U	**0.0062**	**J**	0.011	U	0.011	U
Benz[a]anthracene	0.010	U	0.011	U	0.011	U	0.011	U
Benzo(a)pyrene	0.010	U	0.011	U	0.011	U	0.011	U
Benzo(b)fluoranthene	0.010	U	0.011	U	0.011	U	0.011	U
Benzo(ghi)perylene	0.010	U	0.011	U	0.011	U	0.011	U
Benzo(k)fluoranthene	0.010	U	0.011	U	0.011	U	0.011	U
Carbazole	0.010	UJ	0.028	UJ	0.011	UJ	0.011	UJ
Chrysene	0.010	U	0.011	U	0.011	U	0.011	U
Dibenzo(a,h)anthracene	0.010	U	0.011	U	0.011	U	0.011	U
Dibenzofuran	0.010	UJ	**0.049**	**J**	0.011	UJ	0.011	UJ
Fluoranthene	0.010	U	0.02	U	0.011	U	0.011	U
Fluorene	0.010	UJ	**0.068**	**J**	0.011	UJ	0.011	UJ
Indeno(1,2,3-cd)pyrene	0.010	U	0.011	U	0.011	U	0.011	U
Naphthalene	0.010	U	**0.080**		0.011	U	0.011	U
Phenanthrene	0.010	U	**0.038**		0.011	U	0.011	U
Pyrene	0.010	U	**0.0088**	**J**	0.011	U	0.011	U
Retene	0.010	U	0.011	U	0.011	U	0.011	U
Oxygenated PAHs (ug/L)								
1,4-Anthraquinone	REJ		REJ		REJ		REJ	
4H-Cyclopenta[def]phenanthren-4-one	0.021	U	**0.017**	**J**	0.021	U	0.022	U
5,12-Naphthacenequinone	0.021	UJ	0.022	UJ	0.021	UJ	0.022	UJ

Location	INDIAN GW-1		QUINCE STW		INDIAN GW-2		Equip. Blank	
Sample No.	1304069-01		1304069-02		1304069-03		1304069-04	
Date	4/25/2013		4/26/2013		4/26/2013		4/26/2013	
Time	1500		1425		1535		1700	
7,12-Benz[a]anthracenquinone	0.021	UJ	0.022	UJ	0.021	UJ	0.022	UJ
9,10-Anthracenedione	0.051	U	0.054	U	0.053	U	0.054	U
9,10-Phenthrenequinone	REJ		REJ		REJ		REJ	
9H-Fluoren-9-one	0.051	U	**0.052**	**J**	0.053	U	0.054	U
Aceanthracenequinone	0.051	U	0.054	U	0.053	U	0.054	U
Acenaphthenequinone	0.10	UJ	0.11	UJ	0.11	UJ	0.11	UJ
Benzanthrone	0.021	U	0.022	U	0.021	U	0.022	U
Benzo[a]fluorenone	0.021	U	0.022	U	0.021	U	0.022	U
Benzo[c]phenanthrene-1[1,4]quinone	REJ		REJ		REJ		REJ	
Benzo[cd]pyrenone	0.021	UJ	0.022	UJ	0.021	UJ	0.022	UJ
Phenanthrene-1,4-dione	REJ		REJ		REJ		REJ	

Site Descriptions and Comments:
INDIAN GW-1: Seep adjacent to original downstream trout hatchbox location (I-2C in 2013)
QUINCE STW: Baseflow (groundwater) from suspect stormwater culvert
INDIAN GW-2: Seep 30 ft upstream of original downstream trout hatchbox location (I-2C in 2013)
Equip. Blank: Deionized water from Manchester Laboratory run through Silastic tubing and filter

Bold values indicate detected results
J = Analyte was positively identified; reported result is an approximate concentration
U = Not detected above the reported quantitation limit
UJ = Not detected above the reported estimated quantitation limit
NA = Not analyzed
REJ = Result rejected due to quality control failures

Table E-12. BNA Results and Tentatively Identified Compounds for Groundwater in Lower Indian Creek, ug/L.

Location	INDIAN GW-1		QUINCE STW		INDIAN GW-2		Equip. Blank	
Sample No.	1304069-01		1304069-02		1304069-03		1304069-04	
Date	4/25/2013		4/26/2013		4/26/2013		4/26/2013	
Time	1500		1425		1535		1700	
1,2,4-Trichlorobenzene	0.798	U	0.773	U	0.824	U	0.824	U
1,2-Benzenedicarboxylic acid, bis(2-meth	**0.113**	**NJ**	ND		**0.125**	**NJ**	ND	
1,2-Dichlorobenzene	0.798	U	0.773	U	0.824	U	0.824	U
1,2-Diphenylhydrazine	0.798	U	0.773	U	0.824	U	0.824	U
1,3-Dichlorobenzene	0.798	U	0.773	U	0.824	U	0.824	U
1,4-Benzenedicarboxylic acid, dimethyl e	**0.0787**	**NJ**	ND		**0.123**	**NJ**	ND	
1,4-Dichlorobenzene	0.798	U	0.773	U	0.824	U	0.824	U
2,4,5-Trichlorophenol		REJ	3.09	U	3.3	U	3.3	U
2,4,6-Trichlorophenol	3.19	U	3.09	U	3.3	U	3.3	U
2,4-Dichlorophenol	7.98	U	7.73	U	8.24	U	8.24	U
2,4-Dimethylphenol	7.98	U	7.73	U	8.24	U	8.24	U
2,4-Dinitrophenol	7.98	U	7.73	U	8.24	U	8.24	U
2,4-Dinitrotoluene	3.19	U	3.09	U	3.3	U	3.3	U
2,6-Dinitrotoluene	3.19	U	3.09	U	3.3	U	3.3	U
2-Chlorophenol	3.19	U	3.09	U	3.3	U	3.3	U
2-Cyclohexen-1-Ol	ND		ND		**0.0905**	**NJ**	**0.189**	**NJ**
2-Cyclohexen-1-One	**0.245**	**NJ**	**0.289**	**NJ**	**0.615**	**NJ**	**0.342**	**NJ**
2-Methylphenol	7.98	U	7.73	U	8.24	U	8.24	U
2-Nitroaniline	16	U	15.5	U	16.5	U	16.5	U
2-Nitrophenol	1.6	U	1.55	U	1.65	U	1.65	U
3,3'-Dichlorobenzidine	1.6	U	1.55	U	1.65	U	1.65	U
3B-Coprostanol	7.98	U	7.73	U	8.24	U	8.24	U
3-Cyanocarbazole	ND		**0.0522**	**NJ**	ND		ND	
3-Nitroaniline	3.19	UJ	3.09	UJ	3.3	UJ	3.3	UJ
4,6-Dinitro-2-Methylphenol	16	U	15.5	U	16.5	U	16.5	U
4-Bromophenyl phenyl ether	1.6	U	1.55	U	1.65	U	1.65	U
4-Chloro-3-Methylphenol	7.98	U	7.73	U	8.24	U	8.24	U
4-Chloroaniline		REJ		REJ	33	UJ	33	U
4-Chlorophenyl-Phenylether	0.798	U	0.773	U	0.824	U	0.824	U
4-Methylphenol	7.98	UJ	7.73	U	8.24	U	8.24	U
4-Nitroaniline	3.19	U	3.09	U	3.3	U	3.3	U
4-Nitrophenol	7.98	U	7.73	U	8.24	U	8.24	U
4-nonylphenol	3.19	U	3.09	U	3.3	U	3.3	U
Benzoic Acid	7.98	U	7.73	U	8.24	U	8.24	U
Benzyl Alcohol	7.98	U	7.73	U	8.24	U	8.24	U
Bis(2-chloro-1-methylethyl) ether	0.798	U	0.773	U	0.824	U	0.824	U
Bis(2-Chloroethoxy)Methane	0.798	U	0.773	U	0.824	U	0.824	U

Location	INDIAN GW-1		QUINCE STW		INDIAN GW-2		Equip. Blank	
Sample No.	1304069-01		1304069-02		1304069-03		1304069-04	
Date	4/25/2013		4/26/2013		4/26/2013		4/26/2013	
Time	1500		1425		1535		1700	
Bis(2-Chloroethyl)Ether	1.6	U	1.55	U	1.65	U	1.65	U
Bis(2-Ethylhexyl) Phthalate	**16.3**	**J**	1.55	U	1.65	U	1.65	U
Bisphenol A	3.19	U	3.09	U	3.3	U	3.3	U
Butane, 1,1'-[oxybis(2,1-ethanediyloxy)]	ND		**1.43**	**NJ**	ND		ND	
Butyl benzyl phthalate	3.19	U	3.09	U	3.3	U	3.3	U
Caffeine	1.6	U	1.55	U	1.65	U	1.65	U
Cholesterol	7.98	UJ	7.73	UJ	8.24	UJ	8.24	UJ
cis-13-Octadecanoic acid	ND		**2.22**	**NJ**	ND		ND	
Diethyl phthalate	1.6	U	1.55	U	1.65	U	1.65	U
Dimethyl phthalate	1.6	U	1.55	U	1.65	U	1.65	U
Di-N-Butylphthalate	0.798	U	0.924	U	1.17	U	0.954	U
Di-N-Octyl Phthalate	1.6	U	1.55	U	1.65	U	1.65	U
Dodecane, 1,1'-oxybis-	ND		**0.0602**	**NJ**	ND		ND	
Ethanol, 2-(2-Butoxyethoxy)-	ND		ND		ND		**0.742**	**NJ**
Ethanol, 2-(2-butoxyethoxy)-(1)	ND		**0.0671**	**NJ**	ND		ND	
Ethanol, 2-(2-Butoxyethoxy)-, Acetate	**68.7**	**NJ**	**57.4**	**NJ**	**76.2**	**NJ**	**24.1**	**NJ**
Ethanol, 2,2'-Oxybis-, Diacetate	**0.121**	**NJ**	ND		ND		**0.74**	**NJ**
Hexachlorobenzene	0.798	U	0.773	U	0.824	U	0.824	U
Hexachlorobutadiene	0.798	U	0.773	U	0.824	U	0.824	U
Hexachlorocyclopentadiene	3.19	U	3.09	U	3.3	U	3.3	U
Hexachloroethane	0.798	U	0.773	U	0.824	U	0.824	U
Hexanedioic acid, bis(2-ethylhexyl) este	ND		**0.0677**	**NJ**	ND		ND	
Isophorone	1.6	U	1.55	U	1.65	U	1.65	U
Nitrobenzene	0.798	U	0.773	U	0.824	U	0.824	U
N-Nitrosodi-n-propylamine	0.957	UJ	0.928	UJ	0.989	UJ	0.989	UJ
N-Nitrosodiphenylamine	1.6	U	1.55	U	1.65	U	1.65	U
Nonanal	ND		**0.511**	**NJ**	**0.348**	**NJ**	ND	
Nonanoic Acid	ND		**0.239**	**NJ**	**0.189**	**NJ**	ND	
Octadecanoic Acid	**0.585**	**NJ**	**0.911**	**NJ**	**0.219**	**NJ**	**0.293**	**NJ**
Oleic Acid	**0.589**	**NJ**	ND		**0.517**	**NJ**	**0.783**	**NJ**
Pentachlorophenol	0.798	U	0.773	U	0.824	U	0.824	U
Pentanoic acid, 2,2,4-trimethyl-3-carbox	ND		**0.213**	**NJ**	ND		ND	
Phenol	3.19	U	3.09	U	3.3	U	3.3	U
Phenol, 2,4-bis(1,1-dimethylethyl)-	**0.188**	**NJ**	ND		ND		0.178	
Phenol, 3,5-bis(1,1-dimethylethyl)-	ND		**0.125**	**NJ**	**0.0917**	**NJ**	ND	
Phthalic acid, isobutyl nonyl ester	ND		**0.125**	**NJ**	ND		ND	
Phthalic acid, decyl isobutyl ester	ND		ND		ND		**0.105**	**NJ**
Propanoic acid, 2-methyl-, 1-(1,1-dimeth	ND		ND		**0.104**	**NJ**	**0.0886**	**NJ**
Triclosan	0.798	U	0.773	U	0.824	U	0.824	U

Location	INDIAN GW-1	QUINCE STW	INDIAN GW-2	Equip. Blank
Sample No.	1304069-01	1304069-02	1304069-03	1304069-04
Date	4/25/2013	4/26/2013	4/26/2013	4/26/2013
Time	1500	1425	1535	1700
Triethyl citrate	3.19 U	3.09 U	3.3 U	3.3 U
Unknown Hydrocarbon 24.361	ND	ND	ND	**0.102 NJ**
Unknown Hydrocarbon 25.679	ND	ND	ND	**0.177 NJ**
Unknown Hydrocarbon 25.685	ND	**0.0461 NJ**	ND	ND
Unknown Hydrocarbon 26.304	ND	ND	ND	**0.128 NJ**
Unknown Hydrocarbon 26.908	ND	ND	**0.0524 NJ**	**0.108 NJ**
Unknown Hydrocarbon 27.009	ND	**0.12 NJ**	ND	ND
Unknown Hydrocarbon 28.052	ND	ND	**0.0803 NJ**	ND
Unknown Hydrocarbon 28.608	ND	ND	**0.0598 NJ**	**0.0999 NJ**

Site Descriptions and Comments:
INDIAN GW-1: Seep adjacent to original downstream trout hatchbox location (I-2C in 2013)
QUINCE STW: Baseflow (groundwater) from suspect stormwater culvert
INDIAN GW-2: Seep 30 ft upstream of original downstream trout hatchbox location (I-2C in 2013)
Equip. Blank: Deionized water from Manchester Laboratory run through Silastic tubing and filter

Bolded values indicate detected results
J = Analyte was positively identified; reported result is an approximate concentration
U = Not detected above the reported quantitation limit
UJ = Not detected above the reported estimated quantitation limit
NJ = Analyte was tentatively identified; reported result is an approximate concentration
ND = Not detected (qualitative)
REJ = Result rejected due to quality control failure

Appendix F. Weather during the 2013 Study

Weather data in Table F-1 were accessed from the Weather Underground (www.wunderground.com) for the East Olympia Weather Station.

Table F-1. East Olympia weather April through June 2013.

April	Temperature (° C)			Sunlight		Rain
	high	average	low	watts/m^2	Duration (hours)	cm
15	12.8	6.1	-0.6	303	12:51	0.23
16	14.4	6.7	0.6	341	13:36	0.03
17	15.6	7.2	-0.6	287	13:06	0
18	12.8	8.9	6.1	139	13:21	0.05
19	13.3	10.6	8.3	170	13:06	0.33
20	14.4	10.0	7.2	229	13:50	0
21	13.9	8.9	4.4	212	13:51	0.18
22	16.1	7.8	0.0	417	14:07	0.03
23	17.2	8.3	-1.7	390	13:22	0
24	20.6	11.1	0.6	454	13:51	0
25	24.4	12.8	2.2	406	14:06	0
26	23.9	12.8	5.6	370	13:51	0
27	15.0	11.7	9.4	134	14:05	0
28	15.6	11.1	8.9	261	13:50	0.56
29	13.3	8.9	3.3	459	14:05	0.03
30	13.3	7.8	2.2	354	14:06	0
May	Temperature (° C)			Sunlight		Rain
	high	average	low	watts/m^2	Duration (hours)	cm
1	17.2	8.9	-1.1	465	14:08	0
2	20.6	11.7	2.2	394	14:21	0
3	21.7	12.8	3.3	475	14:21	0
4	26.7	16.7	6.1	483	14:08	0
5	28.9	17.8	5.6	486	14:21	0
6	30.0	18.3	6.7	485	14:37	0
7	21.1	13.9	8.9	366	14:21	0
8	21.1	13.9	8.9	308	14:21	0
9	23.9	13.9	6.7	367	14:20	0
10	27.8	17.2	6.7	432	14:35	0
11	28.9	18.3	10.6	408	14:35	0
12	21.7	17.2	13.9	195	14:05	0.05
13	18.3	13.3	6.7	227	14:36	0.58
14	20.0	12.2	6.1	380	14:51	0
15	16.1	11.1	5.0	133	14:34	0.13

16	20.6	14.4	9.4	329	14:50	0.53
17	18.3	13.3	9.4	278	14:36	0.08
18	15.6	12.2	8.9	164	14:36	0.10
19	18.9	13.3	8.9	207	14:21	0
20	21.1	12.8	6.7	339	14:52	0.08
21	14.4	10.0	7.8	272	14:05	1.24
22	10.0	7.8	5.6	125	15:06	0.66
23	10.0	7.8	5.0	107	14:04	1.02
24	15.0	10.6	7.2	201	14:49	0.48
25	16.1	11.7	8.3	211	14:51	0.05
26	13.3	10.6	9.4	198	14:21	0.15
27	16.1	12.8	10.6	139	14:49	0.97
28	16.7	12.8	10.6	199	14:57	0.10
29	16.7	11.7	8.9	241	15:06	1.17
30	16.1	11.7	7.8	245	15:07	0.10
31	18.9	12.8	7.2	349	15:20	0
June	Temperature (° C)			Sunlight		Rain
	high	average	low	watts/m^2	Duration (hours)	cm
1	22.8	15.6	8.9	391	15:36	0
2	20.0	15.0	11.1	245	15:21	0
3	23.3	14.4	6.7	408	15:36	0
4	27.2	17.2	6.1	502	15:37	0
5	26.7	17.8	9.4	434	15:21	0
6	26.1	17.2	10.0	427	15:36	0
7	21.7	15.0	10.0	254	15:19	0
8	21.7	14.4	7.8	416	15:35	0
9	21.1	13.9	8.3	301	15:37	0
10	21.1	12.2	4.4	351	15:51	0
11	17.8	12.8	7.2	361	15:35	0.03
12	18.3	12.8	8.3	258	15:35	0.20
13	20.0	13.3	10.0	289	15:20	0.38
14	18.9	13.3	9.4	237	15:35	0.03
15	25.0	16.1	6.1	424	15:51	0
16	23.3	16.7	10.0	385	15:51	0
17	23.9	16.7	12.2	366	15:04	0.23
18	21.7	15.0	10.6	366	15:36	0.05
19	21.1	15.0	10.0	361	15:36	0
20	16.7	13.3	10.6	152	15:21	0.03