Effects of treated and untreated stormwater runoff on three benthic macroinvertebrate taxa: mayflies, stoneflies, and caddisflies

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ABSTRACT

Urbanization is accompanied by an increase in impervious surfaces, such as roadways, which are a physical barrier to precipitation infiltration. Stormwater—the water that runs off impervious surfaces after a precipitation event—contains a toxic cocktail of contaminants derived from automobile use. Stormwater can negatively impact stream biota, including fishes and benthic macroinvertebrates. While substantial research has examined the effects of stormwater runoff on Puget Sound coho salmon, stormwater effects on benthic macroinvertebrates are comparatively understudied. I propose a study to explore the effects of stormwater runoff on three benthic macroinvertebrate taxa: mayflies (*Ephemeroptera* spp.), stoneflies (*Plecoptera* spp.), and caddisflies (*Trichoptera* spp.). Under controlled laboratory conditions, I will expose specimens to isolated stormwater contaminants, including polycyclic aromatic hydrocarbons and metals, to determine sensitivity levels. I will also expose specimens to untreated and treated highway runoff to determine whether biofiltration reduces levels of stormwater-induced mortality in benthic macroinvertebrates. Finally, I will field test my laboratory results with stormwater samples and macroinvertebrate richness indices derived from the Puget Sound Stream Benthos database. Data from this study will expand the existing knowledge of stormwater effects on stream biota, and move scientists and managers toward an ecosystem-based approach for Puget Sound stormwater management.

INTRODUCTION

As urbanization encroaches on natural landscapes, the aquatic networks that drain these landscapes are subject to increased pollutant contamination. Urbanized landscapes are characterized by an increase in impervious surfaces, which act as a physical barrier to soil infiltration (Paul and Meyer 2001, Roy et al. 2014, Spromberg et al. 2016). Stormwater—the water that runs off impervious surfaces after a precipitation event—can be a vector for transport of terrestrial pollutants into aquatic ecosystems (Kayhanian et al. 2012). Stormwater runoff is regulated under the National Pollutant Discharge Elimination System, which requires that urban jurisdictions maintain a stormwater management program (US EPA 2010). Management is often achieved with a network of detention and retention ponds that collect stormwater and allow it to infiltrate into the ground. Green stormwater infrastructure (GSI) technologies—such as biofiltration modules, rain gardens, and pervious pavement—have also emerged as a means of decentralized stormwater infiltration and treatment (Roy et al. 2014).

Despite these management strategies, insufficient capacity and high-precipitation events mean that stormwater runoff routinely escapes or evades the infrastructure designed to contain it. When this occurs, stormwater runs downhill and picks up toxic pollutants from road surfaces, which ultimately collect and concentrate in lowland streams (Kayhanian et al. 2008; Roy et al. 2014). The composition of stormwater is complex and highly variable, containing a "cocktail" of contaminants associated with automobile use including byproducts of vehicle exhaust, leaking fluids, wearing tires, and asphalt degradation (Smith and Kaster 1983, Feist et al. 2011, Scholz et al. 2011). Prominent components include petroleum-derived polycyclic aromatic hydrocarbons (PAHs) and metals such as cadmium, copper, nickel, lead, and zinc (Kayhanian et al. 2012, Spromberg et al. 2016). In addition to chemical contamination, stormwater inputs can increase water temperature, decrease dissolved oxygen, and cause bed scour and bank erosion in streams (Seattle Public Utilities 2007; Roy et al. 2014). Together, the combined impacts of stormwater pollution can dramatically alter stream biota and ecosystem processes (Smith and Kaster 1983, Paul and Meyer 2001).

As year-round residents of the stream bed, benthic macroinvertebrates are constantly exposed to stream conditions, including upstream stormwater inputs. Benthic macroinvertebrates play a key trophic role in stream ecosystems: they drive the nutrient recycling process by breaking down organic detritus that falls into the stream, prey upon plankton and other small invertebrates, and provide a food source for higher trophic level organisms (Cummins 1974, Vannote et al. 1980, Nakano et al. 2001). Moreover, due to their continuous exposure to stream conditions, benthic macroinvertebrates are important bioindicators of water quality. While analysis of a water sample can provide a snapshot of water quality at a given moment in time, analysis of the stream's benthic macroinvertebrate assemblage can provide integrated insight into stream conditions year-round (Seattle Public Utilities 2007).

Mayflies (*Ephemeroptera* spp.), stoneflies (*Plecoptera* spp.), and caddisflies (*Trichoptera* spp.) comprise three sensitive orders of larval benthic macroinvertebrates that are central to ecosystem functioning and serve as useful water quality bioindicators (Cummins 1974, Paul and Meyer 2001). Specifically, mayflies are grazers that eat algae, and are particularly intolerant of heavy metal contamination. Caddisflies may be scrapers that collect food from rock surfaces or predators that entrap other small invertebrates in elaborate nets, and are notable for their sensitivity to anthropogenic disturbance (Hachmoller et al. 1991). Stoneflies may be shredders that feed on leaf detritus or predators of other small invertebrates, and require a high-oxygen, low pollution environment (Cummins 1974, King County 2018). These three macroinvertebrate taxa are generally abundant in pristine streams. However, their populations tend to be reduced or absent in urban streams that experience routine flushes of toxic stormwater runoff (Hachmoller et al. 1991, Paul and Meyer 2001, Seattle Public Utilities 2007).

MOTIVATION FOR RESEARCH

A substantial amount of research has been invested in determining the effects of stormwater runoff on Puget Sound salmonids. A primary focus has been the identification of the individual pollutant or "pollutant cocktail" responsible for causing widespread pre-spawn mortality in adult coho salmon (*Oncorhynchus kisutch*). While the specific cause of pre-spawn mortality has yet to be pinpointed, numerous studies point to a chemical component of stormwater runoff as the likely culprit (Scholz et al. 2011; Feist et al. 2011, 2017; McIntyre et al. 2015). In light of this consensus, subsequent research has focused on the efficacy of GSI biofiltration systems in reducing coho pre-spawn mortality (McIntyre et al. 2015, Spromberg et al. 2016).

Relative to the growing body of work on salmon, comparatively little research has been conducted to assess the impacts of stormwater pollutants on benthic macroinvertebrates, nor the effectiveness of biofiltration in ameliorating these impacts. What little research exists is largely comprised of field studies that assess macroinvertebrate diversity and biomass along a stormwater pollution gradient. For example, Pratt (1981) found decreased diversity and abundance of macroinvertebrates—including mayflies and caddisflies—with increasing levels of stormwater runoff. Smith and Kaster (1983), on the other hand, found that physical stream parameters were more predictive of low macroinvertebrate diversity and abundance than were stormwater inputs. More recently, Goldyn et al. (2018) found that macroinvertebrate diversity and abundance were significantly lower at study sites in the vicinity of stormwater outfalls, especially those associated with high levels of car traffic.

While field observations tend to suggest a correlation between stormwater runoff and macroinvertebrate community decline, the literature is lacking in laboratory studies that confirm a causal relationship. Moreover, it is unknown which pollutant or suite of pollutants induces mortality, and at what concentrations mortality occurs. A single laboratory experiment examined effects on untreated runoff on both adult and neonate mayflies (*Baetis* spp.). Findings revealed that stormwater exposure severely impaired mayfly survival and reproduction; however, sensitivity to individual stormwater components such as PAHs and heavy metals was not explored (McIntyre et al. 2015). Subsequent tests exposing mayflies to stormwater treated with a simple biofiltration mixture of compost, sand, and bark resulted in substantially lower mortality rates but did not eliminate stormwater-induced mortality completely, whereas coho salmon exposed to the same treated stormwater experienced no mortality (McIntyre et al. 2015, Spromberg et al. 2016). These results indicate that mayflies may exhibit heightened sensitivity to toxic stormwater runoff relative to coho salmon. Alternatively, mayflies may be sensitive to a particular stormwater pollutant that is not fully removed with biofiltration. No comparable controlled laboratory studies have examined the impacts of stormwater pollutants on other key macroinvertebrate taxa, including stoneflies and caddisflies.

In short, the efficacy of GSI biofiltration systems that eliminate pre-spawn mortality in coho salmon is underexplored with respect to benthic macroinvertebrates. In contrast to the controlled laboratory experiments on mayflies by McIntyre et al. (2015), field tests found no significant changes to the benthic macroinvertebrate assemblage within a particular catchment basin following installation of rain barrels and rain gardens (Roy et al. 2014). As local jurisdictions grapple with how to manage stormwater inputs to local streams, it is imperative that we understand whether the proposed treatment options will reduce stormwater-induced mortality in not just coho salmon but all key stream biota, including benthic macroinvertebrates.

RESEARCH QUESTIONS

1. How does exposure to various combinations of stormwater pollutants impact the survival of three benthic macroinvertebrate taxa: mayflies, stoneflies, and caddisflies?
2. Does biofiltration reduce or eliminate stormwater-induced mortality in mayflies, stoneflies, and caddisflies?
3. Are the stormwater pollutant impacts observed a laboratory setting replicated in the field, as evidenced by macroinvertebrate taxa richness at a variety of representative sampling sites?

HYPOTHESES

1. In general, higher concentrations of PAHs and metals in artificial stormwater will produce higher levels of mortality in mayflies, stoneflies, and caddisflies. All benthic macroinvertebrate taxa will exhibit highest levels of mortality when exposed to runoff that contains both PAHs and heavy metals, due to additive effects of the combined contaminants. Different taxa will demonstrate varying levels of sensitivity to artificial stormwater formulations that isolate particular contaminants.
2. Biofiltration will produce different results for mayflies, stoneflies, and caddisflies. In general, biofiltration is expected to reduce, but not eliminate, stormwater-induced mortality in all taxa. Mayflies are expected to see the greatest reduction in mortality, followed by caddisflies.

Stoneflies, as the most sensitive taxa considered, are expected to see some reduction in mortality, but less so than mayflies and caddisflies.

3. In the field, benthic macroinvertebrate taxa richness will decrease along an urban gradient. For sites with high levels of stormwater contaminants, benthic macroinvertebrate taxa richness will be lower for a given stormwater contaminant level relative to that predicted by laboratory results. This is because urban streams are generally degraded by additional physical factors, such as increased temperature and sedimentation, which further reduce their suitability as habitat for mayflies, stoneflies, and caddisflies.

EXPERIMENTAL DESIGN AND METHODS

Laboratory study

Live macroinvertebrate specimens will be collected from the Cedar River, a relatively pristine system protected as a drinking water source for the Seattle metropolitan area. Specimens will be collected using a D-frame kick net and transported back to the lab in aerated river water. Macroinvertebrates will be sorted by taxa (mayfly, stonefly, caddisfly) and ontogeny (neonate, adult) and randomly placed in experimental chambers in groups of ten (McIntyre et al. 2015). Environmental chambers will contain one of five stormwater treatments applied in either low or high concentrations, mimicking the stormwater toxicity tests conducted on coho salmon by Spromberg et al (2016; Table 1). The control chamber will contain water collected from the Cedar River at the site of specimen collection, and all treatments will use Cedar River water as a solvent. Macroinvertebrate specimens will be exposed to stormwater treatments for 48 hours, followed by exposure to control water for 5 days. Observations will occur at 24-hour intervals to quantify mortality; dead specimens will be removed from environmental chambers.

Table 1. Stormwater treatments to which mayfly, stonefly, and caddisfly specimens will be exposed. Artificial treatments mimic pollutant combinations and concentrations found at or above those measured in urban streams following autumn storm events. Highway runoff formulations will be collected directly from downspouts on the WA-520 bridge.

Treatment	***Pollutants***	***Solvent***
Control (Cedar River)	N/A	N/A
Artificial stormwater (PAHs)	Alaskan North Slope crude oil Pyrogenic pyrene Fluoranthene	Cedar River water
Artificial stormwater (Metals)	Cadmium ($CdCl_2$) Nickel ($NiCl_2$) Lead ($PbCl_2$) Copper ($CuCl_2$) Zinc ($ZnCl_2$)	Cedar River water
Artificial stormwater (PAHs + Metals)	Combination of PAH and metals pollutants	Cedar River water
Highway stormwater runoff (untreated)	Pending analysis by external water quality laboratory	Pending analysis by external water quality laboratory
Highway stormwater runoff (treated)	Pending analysis by external water quality laboratory	Pending analysis by external water quality laboratory

Field study

Water samples will be collected from streams in the Puget Sound watershed within one hour of a local precipitation event and analyzed for concentrations of PAHs and metals (cadmium, nickel, lead, copper, zinc). Sampling sites will be selected from the existing database of Puget Sound Stream Benthos (PSSB) monitoring locations (King County, 2018; www.pugetsoundstreambenthos.org). Only sites with at least contiguous ten years of macroinvertebrate monitoring data will be considered for analysis. The Nature Conservancy's

Solving Stormwater model will be used to select sites that represent a mix of land uses (open space, residential, commercial, industrial) and estimated stormwater pollutant loads (The Nature Conservancy 2017; igeowater.com/dev/tnc/#/).

Following water quality analysis, the stormwater contaminant composition of each water sample will be correlated with macroinvertebrate taxa richness for the corresponding site. Stormwater samples will be provided to an external water quality laboratory for determination of PAH and metals levels. Macroinvertebrate richness will be determined using Benthic Index of Biotic Integrity (B-IBI) scores recorded in the PSSB database, which provide a quantitative ranking of the biological condition of Puget Sound streams. The B-IBI score for a given stream site is predicated, in part, on the abundance of mayfly, stonefly, and caddisfly taxa in that stream relative to what would be expected at a pristine Puget Sound stream. The presence or absence of particular benthic macroinvertebrate taxa with respect to the stormwater contaminant levels of each stream site will be used as an indicator of taxa tolerance and intolerance, respectively. These data will be compared with the results of our laboratory experiments and used to determine whether mortality-inducing contaminant effects observed in a controlled laboratory setting are reflected by the absence of particular taxa in the field.

ANTICIPATED RESULTS

I expect that exposure to artificial stormwater treatments containing PAHs, metals, or a combination of both will cause mortality in mayflies, stoneflies, and caddisflies (Figure 1). With respect to individual treatments, I expect that the mixture of PAHs and metals will produce the highest levels of mortality in all three taxa, due to additive effects of the combined contaminants. Because the three benthic macroinvertebrate taxa have known sensitivities to different contaminants, mortality levels are expected to vary between them. In particular, mayflies are likely to have higher mortality than stoneflies and caddisflies when exposed to the metals treatment due to their known metals intolerance (King County 2018).

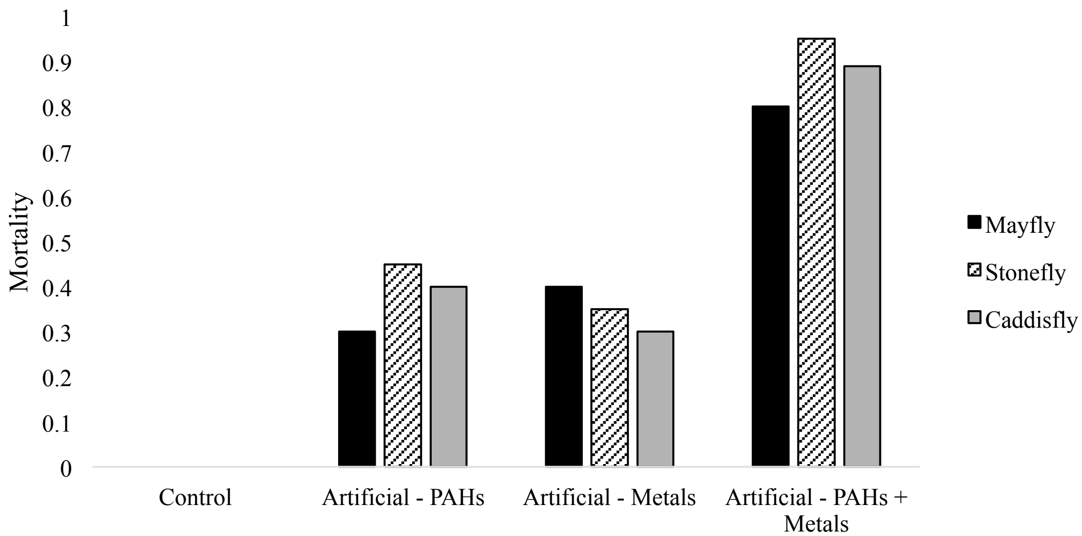

Figure 1. Mortality in mayflies, stoneflies, and caddisflies after 48-h exposure to a variety of artificial stormwater treatments followed by 5 days of clean water exposure

When exposed to untreated and treated stormwater runoff from the WA-520 bridge, I expect that untreated runoff will cause higher levels of mortality than treated runoff (Figure 2). As observed by McIntyre et al. (2015) with respect to mayflies, I expect that biofiltration will remove some—but not all—of the stormwater contaminants than cause mortality in the three benthic macroinvertebrate taxa. In accordance with the anticipated results of exposure to artificial stormwater formulations, I expect mortality rates to vary between taxa, with stoneflies and caddisflies exhibiting higher sensitivity than mayflies (Hachmoller et al. 1997).

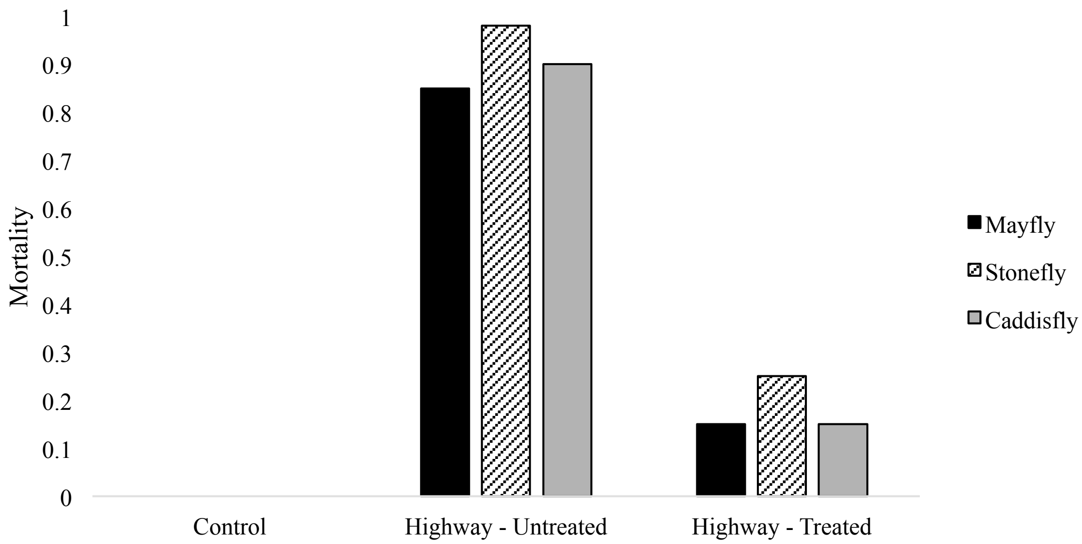

Figure 2. Mortality in mayflies, stoneflies, and caddisflies after 48-h exposure to untreated and treated stormwater runoff from the WA-520 bridge followed by 5 days of clean water exposure.

Several studies have observed a decrease in macroinvertebrate diversity and abundance along an increasingly urban gradient (Pratt 1981, Roy et al. 2014, Goldyn et al. 2018). Accordingly, I expect that taxa richness at open space sites with relatively little stormwater impact will be higher relative to urban sites, with lowest taxa richness at industrial-type sites (Figure 3). Because stoneflies and caddisflies are highly sensitive to anthropogenically disturbed environments that produce increased stream temperatures and low levels of dissolved oxygen, I expect the richness of these taxa to be slightly lower than mayfly taxa richness along an urban gradient.

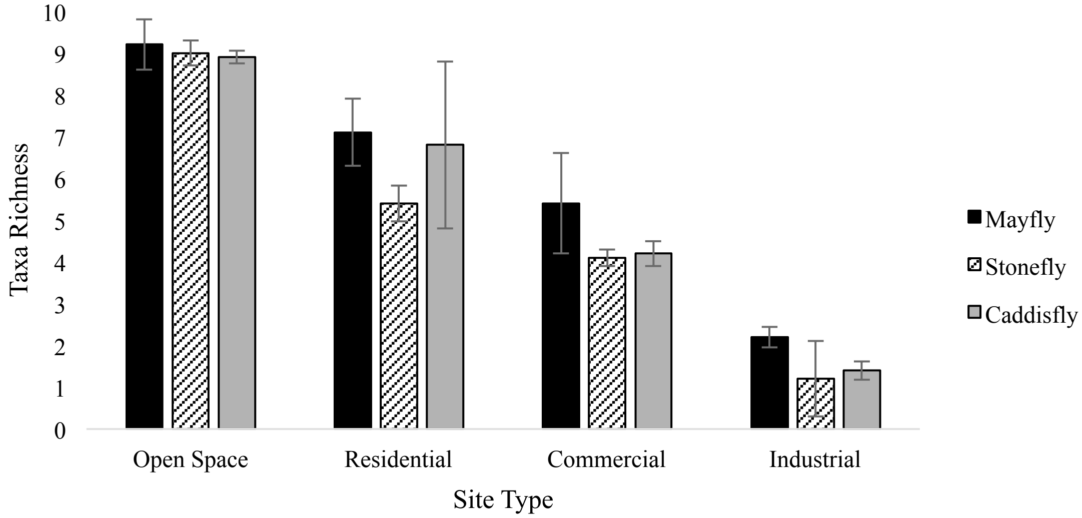

Figure 3. Average taxa richness (+/- SD) of mayfly, stonefly, and caddisfly assemblages from field sites along an urban gradient. Four sites were sampled for each site type. Taxa richness was calculated per Puget Sound Stream Benthos database guidelines (King County 2018).

DISCUSSION

Laboratory tests that expose mayflies, stoneflies, and caddisflies to various artificial stormwater treatments are expected to cause elevated mortality rates relative to control treatments. Mortality data with respect to PAHs, metals, and combined PAHs-metals treatments will indicate whether macroinvertebrate taxa are more sensitive to one suite of stormwater contaminants over another. If, as expected, mortality rates for the combined PAHs-metals treatment exceed the combined mortality rates from treatments that isolate PAHs and metals, additive properties are likely at play that increase stormwater toxicity to benthic macroinvertebrates. We may also detect variation amongst taxa in sensitivity to particular contaminants (Hachmoller et al. 1991, Kayhanian et al. 2008). If some taxa are relatively unaffected by contaminant exposure, it may suggest a competitive advantage over those that are more severely impacted. It is also possible that mortality rates will be relatively uniform across benthic macroinvertebrates. This result would reinforce the need for stormwater treatment solutions designed to treat the complete contaminant profile, as opposed to those designed to remove particular PAHs or metals (Kayhanian et al. 2012).

Following exposure to untreated and treated runoff from the WA-520 bridge, macroinvertebrates exposed to treated stormwater may experience significantly lower rates of mortality relative to those exposed to untreated stormwater. This result would indicate that GSI biofiltration may be an effective solution for reducing stormwater impacts and improving stream habitat, supporting the findings by McIntyre et al. (2015) and Spromberg et al. (2016) pertaining to coho salmon. On the other hand, if treatment does not substantially reduce mortality rates relative to untreated runoff, or if it reduces mortality rates in some macroinvertebrate taxa but not others, this will indicate that the proposed biofiltration system requires further refinement. In this case, laboratory findings regarding the sensitivity levels of mayflies, stoneflies, and caddisflies to PAHs and metals contaminants will inform future biofiltration trials.

Findings derived from field sampling will serve to either corroborate or complicate laboratory findings. Mayfly, stonefly, and caddisfly taxa richness scores from the PSSB database will be compared to stormwater contaminant levels for each field site, and taxa present will be assumed tolerant to the existing contaminant loads. These inferred tolerances will be used to field-truth our laboratory findings. Taxa that survive exposure to a particular stormwater treatment in a controlled laboratory setting are expected to be present at field sites where those contaminants are present, whereas taxa that experience mortality in response to particular stormwater contaminants are expected to be absent.

Accordingly, if the taxa present at field sites along an urban gradient reflect lab findings, this would suggest that stormwater inputs may indeed play a key role in structuring a stream's benthic macroinvertebrate assemblage. In this case, the results of the biofiltration portion of the study are particularly important, as effective treatment of stormwater runoff could result in the recuperation of diminished macroinvertebrate populations in urban streams. Alternatively, if fewer taxa are present at a given stream site relative to what lab findings would suggest, there may be other factors exacerbating benthic macroinvertebrate declines. Potential factors may include physical characteristics of the stream that further impair habitat quality for benthic macroinvertebrates, including elevated sediment loads and warmer thermal regimes common to urban streams (Smith and Kaster 1983, Roy et al. 2014). In this case, future work should focus on the impacts of multiple stressors on benthic macroinvertebrates. That said, if more taxa are present at field sites relative to what lab findings would suggest, there may be environmental factors acting to mitigate the impacts of stormwater runoff of benthic macroinvertebrates. While this would be considered a positive outcome, it also points to a need for additional research to identify and better understand the mechanisms behind these mitigating factors.

Broadly, this suite of laboratory and field studies will contribute to a deeper, more nuanced understanding of the effects of stormwater contaminants on key benthic macroinvertebrate taxa, and how simple solutions like biofiltration may be implemented to mitigate stormwater impacts. Extending the existing literature beyond coho salmon to include other key stream biota, such as benthic macroinvertebrates, will move scientists and managers toward a more comprehensive, ecosystem-based approach for stormwater management and habitat restoration in Puget Sound.

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