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Soil bioretention protects juvenile salmon and their prey from the toxic impacts of urban stormwater runoff

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HIGHLIGHTS

- Characterized the chemistry and toxicity of highway runoff from six storm events.
- Highway runoff caused lethal and sublethal toxicity in juvenile salmon and their prey.
- We treated highway runoff via infiltration through a bioretention soil media (BSM).
- BSM was 60% sand: 15% compost: 15% shredded bark: 10% water treatment residuals.
- Bioretention treatment of runoff prevented all mortality and sublethal toxicity.

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ABSTRACT

Green stormwater infrastructure (GSI), or low impact development, encompasses a diverse and expanding portfolio of strategies to reduce the impacts of stormwater runoff on natural systems. Benchmarks for GSI success are usually framed in terms of hydrology and water chemistry, with reduced flow and loadings of toxic chemical contaminants as primary metrics. Despite the central goal of protecting aquatic species abundance and diversity, the effectiveness of GSI treatments in maintaining diverse assemblages of sensitive aquatic taxa has not been widely evaluated. In the present study we characterized the baseline toxicity of untreated urban runoff from a highway in Seattle, WA, across six storm events. For all storms, first flush runoff was toxic to the daphniid *Ceriodaphnia dubia*, causing up to 100% mortality or impairing reproduction among survivors. We then evaluated whether soil media used in bioretention, a conventional GSI method, could reduce or eliminate toxicity to juvenile coho salmon (*Oncorhynchus kisutch*) as well as their macroinvertebrate prey, including cultured *C. dubia* and wild-collected mayfly nymphs (*Baetis* spp.). Untreated highway runoff was generally lethal to salmon and invertebrates, and this acute mortality was eliminated when the runoff was filtered through soil media in bioretention columns. Soil treatment also protected against sublethal reproductive toxicity in *C. dubia*. Thus, a relatively inexpensive GSI technology can be highly effective at reversing the acutely lethal and sublethal effects of urban runoff on multiple aquatic species.

1. Introduction

Polluted stormwater runoff is one of the most important threats to water quality in the developed and developing world. Green stormwater infrastructure (GSI), also known as low-impact development, encompasses a set of evolving technologies designed to mimic the hydrologic and filtration capacity of undeveloped landscapes. Examples include green roofs, bioretention systems, and permeable pavement (Dietz, 2007; Ahiablame et al., 2012). The overarching aim of GSI is to slow, spread, and infiltrate stormwater runoff in the urban environment, thereby improving water quality and reducing risks to public safety from flooding and combined sewer overflows. In urbanized areas of the United States, the National Pollutant Discharge Elimination System (NPDES) regulates the release of potentially toxic stormwater runoff. To meet permit requirements, municipalities are increasingly incorporating the use of GSI to reduce runoff pollution to waterways (US EPA, 2010).

Urban runoff impacts the hydrology, geomorphology, and thermal regime of urban streams (Paul and Meyer, 2001; Sheeder et al., 2002; Konrad et al., 2005; Kinouchi et al., 2007). Runoff also trans-

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ports chemical contaminants to receiving waters, many of which are toxic to fish (Skinner et al., 1999; Kayhanian et al., 2008; Corsi et al., 2010), invertebrates (Hall and Anderson, 1988; Marsalek et al., 1999; Kayhanian et al., 2008; Corsi et al., 2010), and aquatic plants (Kayhanian et al., 2008). Field assessments have helped disaggregate the impacts of stormwater quantity and quality on stream taxa. This includes, for example, the role of water quality in the recurrent die-offs of adult coho salmon returning to spawn in urban watersheds in western North America (Scholz et al., 2011). More generally, however, it remains difficult to attribute poor indices of biological integrity to degraded water quality versus other known and important drivers of the urban stream syndrome such as increased flow volume and rates (Roy et al., 2003; Morgan and Cushman, 2005; Walsh et al., 2005, 2007).

The identification of inexpensive, scalable GSI technologies that improve water quality and protect or restore aquatic communities is a key goal in the evolving science of stormwater management. The conventional metrics for success are typically reduced surface flows (DeBusk et al., 2011) and contaminant removal, including metals (Davis et al., 2003) and polycyclic aromatic hydrocarbons (PAHs) (LeFevre et al., 2012). Water quality treatment through physical filtration, sorption, and other soil chemistry mechanisms is a common GSI tenet, and green roofs, rain gardens, and bioretention planters are all examples of GSI practices that use soil mixes of different compositions as treatment media. Moreover, soil mixes may or may not be planted with vegetation to further extract excess nutrients or other pollutants (Hunt et al., 2012). The capacity for soil systems to retain or adsorb toxics in runoff is well established. For example, high removal efficiencies have been documented for metals (Davis et al., 2009), PAHs (DiBlasi et al., 2009), oil (Chapman and Horner, 2010), pesticides (Zhang et al., 2010), and nutrients (Davis et al., 2006). By contrast, the effectiveness of soil bioretention mechanisms as a means to reduce or eliminate adverse health effects to aquatic species has not been widely studied.

Here we characterized the toxicity of first flush runoff collected from an urban arterial in Seattle (Washington, USA) across six distinct storm events. This source of stormwater contains pollutants that are typical for roadways with a relatively high motor vehicle traffic volume year-round. Baseline toxicity was measured in terms of the survival and reproductive success of the cladoceran *Ceriodaphnia dubia* – a common model invertebrate in toxicology. For the final storm, we assessed the effectiveness of one GSI technique by filtering highway runoff through large experimental bioretention columns. In addition to survival and reproduction in *C. dubia*, we monitored the survival of juvenile coho salmon (*Oncorhynchus kisutch*) and wild mayfly nymphs (*Baetis* spp.) exposed to GSI-treated and untreated stormwater. Salmon are a keystone species of temperate coastal regions and mayfly nymphs are an important prey item for juvenile salmon as well as many other species.

2. Methods

2.1. Highway runoff collection

Stormwater was collected during six distinct storms between August 2011 and September 2012, following antecedent dry periods (ADP) of 5–50 d. Runoff was captured at NOAA's Northwest Fisheries Science Center (NWFSC; Seattle, WA, USA) from downspouts draining a busy elevated urban highway (annual average daily traffic = 94 000 vehicles in 2011, 67 000 in 2012; WA DOT, 2012). A diverter (Rain Harvesting, AquaBarrel, Gaithersburg, MD, USA) collected the first flush into glass carboys. Coarse detritus was pre-filtered with fiberglass window screen to prevent clogging the intake. Runoff from each storm event was frozen (−20 °C) within 4 h of collection, a procedure that did not alter the toxicity of samples (McIntyre et al., 2014).

The sixth storm (September 2012) was a source of stormwater runoff for the bioretention treatment. Collected runoff (250 L) was transported on ice from Seattle to the Washington State University Research and Extension Center in Puyallup, WA (WSU-P). A larger volume of runoff could not be collected due to the small size of the storm (0.3 mm). The collected sample was therefore diluted to a total volume of 410 L with rainwater collected at WSU-P to achieve sufficient volume for juvenile coho salmon exposures. For experiments with salmon, the runoff (untreated and treated) was used on the day of collection. For experiments with macroinvertebrates, treated and untreated runoff samples were stored in amber glass bottles at −20 °C and thawed at room temperature on the day of use. Carboys were scrubbed with hot water and rinsed with acetone and methylene chloride between storm events.

2.2. Bioretention treatment

Runoff collected during the September 2012 storm event was filtered through experimental soil columns at WSU-P as previously described (McIntyre et al., 2014). Briefly, the stormwater was transferred from glass collection carboys to a high-density polyethylene cistern for homogenization. Pre-treatment runoff was then sampled, and the remaining water in the cistern was filtered through 12 soil bioretention columns (22 L each) at a rate of 0.058 mm s^{-1}. Each column (36 cm diameter) contained a 61 cm deep mixture of 60% sand, 15% compost, 15% shredded bark, and 10% drinking water treatment residuals (City of Anacortes, WA) overlying a 30-cm deep gravel aggregate drainage layer (Palmer et al., 2013). Half of the columns were planted in November 2011 with the sedge *Carex flacca*, while the other half had no plants (Fig. 1). Treated effluent (19 L from each column) was composited by treatment into glass aquaria prior to water sample collection, resulting in three replicates of each water treatment: untreated runoff (Runoff), bioretention with soil only (No Plants), and bioretention with both soil and plants (Plants). An additional three aquaria were filled with WSU-P fish lab water (described below) as a negative control ('Control'). Exposure waters for each

Fig. 1. The two sets of bioretention columns used to filter runoff from the September 2012 storm event. All columns contain a mixed layer of bioretention soil medium overlying a gravel aggregate drainage layer. The columns on the left are planted with *Carex flacca*.

treatment were collected and chemically analyzed as previously reported (McIntyre et al., 2014). Additionally, water chemistry for the Control treatment (juvenile coho and *Baetis* tests) is described in Table S1, Figs. S1, and S2. Within each treatment, composite samples across triplicate aquaria were frozen at −20 °C in amber glass bottles for subsequent biological analyses with *C. dubia* and *Baetis* spp. The remaining water in each aquarium was used immediately for juvenile coho exposures.

2.3. Baseline toxicity of untreated stormwater to C. dubia

To assess the toxicity of untreated highway runoff across multiple storms, cladocerans (*C. dubia)* were cultured at WSU-P as previously described (Deardorff and Stark, 2009). Exposures to runoff or reconstituted de-ionized water (controls) were carried out in glass beakers maintained in an environmental chamber (25 °C; 50% relative humidity; 18:6 h light:dark photoperiod). For exposures lasting 48 h, 10 neonates (<24 h old) were placed in each of four replicates of 30 mL, fed 0.2 mL of food solution at test onset to improve control survival, and counted at 48 h. The significance of *C. dubia* 48 h replicate survival relative to unexposed controls was determined by *t*-test for each runoff event. Reproductive success was measured following longer (7 d) exposures. For each of the five storms, 10 neonates were placed in each of four replicates of 100 mL and received 1 mL of food solution daily. On day 7, the survival of females and the number of offspring per replicate were counted relative to controls. Statistical significance was determined using multivariate general linear models (GLM) with numbers of adults and neonates per adult as dependent variables. All statistics were performed with SPSS v. 21 software (IBM) with an $\alpha = 0.05$. Individuals were added in groups of ten to each treatment by replicate number (i.e., Control 1, Exposed 1, Control 2, Exposed 2, etc.) and were assessed in the same order.

2.4. Toxicity to invertebrates and juvenile salmon pre- and post-soil infiltration

The survival and reproductive success of cladocerans before and after infiltration of stormwater through bioretention columns was assessed using runoff from the September 2012 storm event and significance tested by ANOVA (48 h survival) and multivariate GLM (7 d survival and neonates per adult) with Dunnett post hoc tests. One neonate (<24 h) was placed in each of ten 50 mL glass beakers containing 30 mL of solution and fed 0.2 mL of food solution daily as per U.S. Environmental Protection Agency guidelines for reproductive toxicity testing (U.S. EPA, 2002a). Neonate survival and reproduction was monitored daily for 7 d. Control replicates met minimum survival, brood number, and offspring counts (U.S. EPA, 2002a).

Wild mayfly nymphs, an important source of prey for juvenile salmon, were collected from the protected and nearly pristine Cedar River near Landsburg, WA. Live benthic macroinvertebrates were capture by kicknet and transported in aerated river water to WSU-P. Individual mayflies (*Baetis* spp.) were isolated and placed in groups of 10 in each of six replicate chambers by replicate number (i.e. Control 1, Exposed 1, Control 2, Exposed 2, etc). Chambers consisted of 250 mL Erlenmeyer flasks lined with clean river stones and containing 100 mL of exposure solution. Each flask was capped and fitted with an aeration tube. Flasks were suspended in a cold water bath at 13 °C on a 12:12 light:dark regime. Surviving individuals were counted at 48 h, and replicate survival relative to controls was analyzed by one-way ANOVA with a Dunnett post hoc. Error bars in all figures are one standard error (SE) of the mean unless noted as standard deviation (SD).

Juvenile coho salmon were obtained from the hatchery facility at the NWFSC (Seattle, WA) and maintained at WSU-P in flow-through circular fiberglass tanks supplied with dechlorinated city water at 13 °C on a 12:12 light:dark regime. The subyearling coho ($\bar{x}$, SD: length = 70, 10 mm; weight = 2.8, 1.2 g) were exposed to untreated and treated stormwater (or hatchery control water) in 35-L glass aquaria supplied with an airstone and maintained at 13 °C using a water bath. Aquaria were randomly assigned to water baths. Per U.S. Environmental Protection Agency guidelines for acute toxicity testing (U.S. EPA, 2002b), ten coho were sequentially placed in each aquarium. The pH and dissolved oxygen (DO) measurements at the outset of each test were within a normal range for maintaining healthy juvenile coho (pH = 7.16–7.85, DO = 7.36–9.13 mg L^{-1}).

Dissolved oxygen levels in both the untreated and treated runoff declined over the first 12 h to <6 mg L^{-1}. By this time, 100% of the juvenile coho exposed to untreated runoff had died (vs. 0% in the treated runoff). We therefore placed a new set of live fish in the untreated exposure aquaria and increased aeration in all aquaria. Dissolved oxygen, measured daily, remained within the recommended range for the remainder of the 96 h test (DO = 7.89–10.10 mg L^{-1} at test termination). Survival was monitored daily. Surviving coho were euthanized after 96 h in MS-222. Individual lengths and weights were measured and bile was collected by puncturing the gall bladder with a solvent-rinsed scalpel. Bile was stored at −20 °C until analysis for PAH metabolites at NWFSC (Seattle, WA) using high performance liquid chromatography with fluorescence detection (HPLC-F) (Yanagida et al., 2012), as detailed in Supporting Information (Text S1). Due to the small volume of bile per fish, triplicate samples could only be acquired for one treatment (Plants). Bile from the remaining fish was composited into one replicate per treatment. These unique values (No Plants and Control) for each PAH metabolite (naphthalene, phenanthrene, pyrene, benzo[a]pyrene) were compared to a range of 3 ∗ SD (standard deviation) of the mean values for Plants. Because 3 ∗ SD should encompass 99% of the actual distribution, values beyond this range were assumed to be from a different distribution. Gill tissue, sampled with Teflon scissors and plastic forceps, was composited across replicates in plastic Whirl-paks and stored at −20 °C until metals analysis at Trace Elements Research Laboratory (College Station, TX) by ICP-MS, as detailed in Supporting Information (Text S2). Differences in metal concentration among treatments were analyzed by multivariate GLM.

3. Ethics Statement

Juvenile coho salmon were maintained and euthanized following protocol #00435-001 approved by the Institutional Animal Care and Use Committee at Washington State University. Mayflies were collected under a scientific collection permit issued by the Washington State Department of Fish and Wildlife (Permit #12-250). Humane euthanization was not employed prior to the end of the study because mortality was an important endpoint for each test. Animals were monitored daily. Surviving animals were euthanized at the end of each study by MS-222 overdose (coho salmon), submersion in 80% ethanol (mayfly nymphs), or freezing (*C. dubia*). A total of 150 coho salmon were used, 240 mayflies, and 790 *C. dubia*.

4. Results

4.1. Biological effectiveness of bioretention: macroinvertebrates

Highway runoff caused acute mortality (Fig. 2) and reproductive impairment (Fig. 3) in *C. dubia*. At the end of the 48-h exposure, mortality was significant for five of the six storms (Fig. 2); specifically August 2011 ($t(4) = 7.0$, $p = 0.002$), October 2011

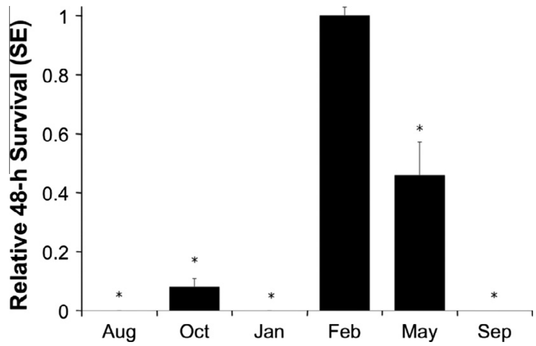

Fig. 2. Survival of *C. dubia* following 48-h exposure to first flush highway runoff relative to control survival for each storm event tested. Asterisks indicate runoff exposures that significantly affected survival relative to controls.

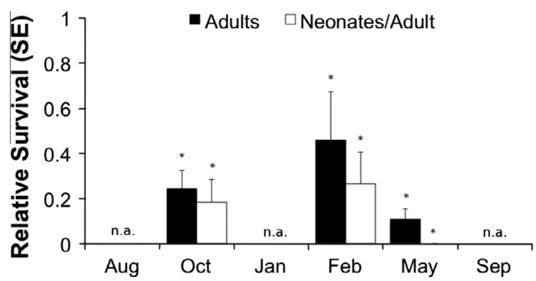

Fig. 3. Survival and reproductive impairment for *C. dubia* following 7-d exposure to highway runoff relative to control values for each storm event tested. Survival and reproduction were impaired following exposure to all runoff samples. n.a. indicates runoff that caused 100% mortality before *C. dubia* reached reproductive maturity.

($t(6) = 10.752$, $p < 0.001$), January 2012 ($t(6) = 12.333$, $p < 0.001$), May 2012 ($t(6) = 3.922$, $p = 0.008$), and September 2012 ($t(6)$ = undefined; 100% mortality in runoff vs. 0% mortality in controls), but not February 2012 ($t(6) = 0$, $p = 1.000$). For storms with survival beyond 48 h, the 7 d test indicated mortality was significant after 7 d in all remaining untreated runoff (Fig. 3); specifically October 2011 ($F(1,1) = 78.400$, $p < 0.001$), February 2012 ($F(1,1) = 6.316$, $p = 0.046$), and May 2012 ($F(1,1) = 289.00$, $p < 0.001$). For the August 2011 event (48 h survival assessment), mortality declined from 100% in the fresh sample to 0% after cold storage (4 °C) in the dark for 7 d. Untreated September 2012 highway runoff was also acutely lethal to wild mayflies (82% mortality; Fig. 4).

Reproduction of *C. dubia* was impaired after 7-d exposure for all runoff events (Fig. 3). No neonates were produced when stormwater treatments resulted in 0% female survival (August 2011, January 2012, September 2012 runoff), but in exposures with

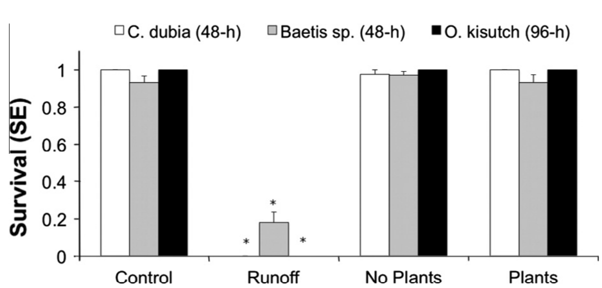

Fig. 4. Survival of three test organisms exposed to control water, untreated September 2012 runoff, runoff treated with bioretention without plants (No Plants), and runoff treated with bioretention with plants (Plants). Asterisks indicate survival significantly lower than control. Error bars are ± one standard error of the mean.

surviving females, significantly fewer offspring were produced per female in runoff compared to control water (October 2011 ($F(1,1) = 62.837$, $p = 0.001$), February 2012 ($F(1,1) = 16.435$, $p = 0.007$), and May 2012 ($F(1,1) = 30.082$, $p = 0.002$).

For daphniids and mayflies, treatment with bioretention (with or without plants) conferred complete protection against the lethal toxicity of stormwater runoff, with survival rates that were not significantly different from controls (Fig. 4). Although *C. dubia* neonate production in bioretention-treated waters initially lagged behind controls (Fig. 5), production was not significantly different from controls by the end of 7 d ($F(3,39) = 37.674$) for daphniids exposed to treated runoff in bioretention with plants (Dunnett post hoc, $p = 0.980$) or without plants ($p = 0.949$).

4.2. Biological effectiveness of bioretention: juvenile salmon

Untreated highway runoff was acutely lethal to juvenile coho salmon (*O. kisutch*), with 100% mortality occurring within 12 h of exposure, with or without dissolved oxygen supplementation. As with the macroinvertebrates, treatment of runoff through the bioretention soil medium prevented mortality (Fig. 4).

Because coho salmon exposed to untreated runoff did not survive to test termination, bile was not collected for analysis of PAH exposure. The bile metabolites measured in surviving salmon from the post-filtration treatments (96-h exposures) were modestly higher than controls, as indicated by being more than 3 SD of the mean (Fig. 6). This indicates that PAHs were still bioavailable in the water passing through the soil columns. The presence of plants in the soil columns reduced the naphthalene-equivalent

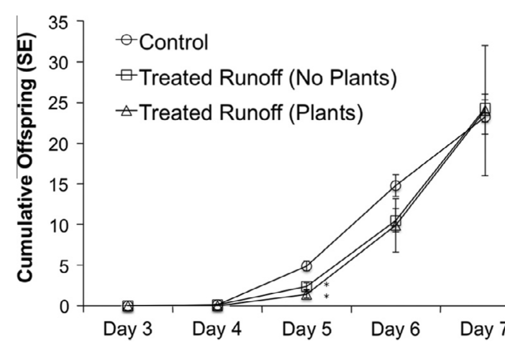

Fig. 5. Cumulative neonate count during the 7-d exposure of *C. dubia* to September 2012 runoff treated with bioretention compared with controls. Neonate production began on Day 4, was significantly lower on Day 5 for treated water exposures, but was not different among treatments by the end of the test period.

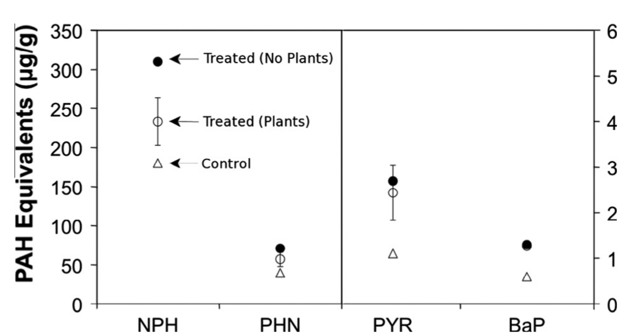

Fig. 6. Biliary metabolites of PAHs in juvenile coho salmon at the end of the 96-h exposure to control water or runoff treated with bioretention, with and without plants. Equivalents of PAHs are for napthalene (NPH), phenanthrene (PHN), pyrene (PYR), and benzo(*a*)pyrene (B*a*P). The error bars on the 'Plants' treatment are ±3 ∗ SD of the mean for the three replicates. Control and 'No Plants' are composite samples of all replicates for those treatments.

Table 1
Mean metal concentrations (mg/kg wet weight) in triplicate samples of gills of juvenile coho salmon exposed to control water or highway runoff treated with bioretention, without plants or with plants.

Mean	Zn	Cu	Cd	Ni	Pb
Control	340	4.473	1.527	<0.502[a]	<0.502
No Plants	383	4.805	2.160	<0.508	<0.508
Plants	320	3.840	1.540	<0.502	<0.502
Std. Dev.[b]					
Control	43	0.568	0.208	n.a.[c]	n.a.
No Plants	13	0.445	0.085	n.a.	n.a.
Plants	47	1.039	0.693	n.a.	n.a.

[a] '<' Indicates value less than detection limit.
[b] One standard deviation of the mean.
[c] Not applicable since below detection limit.

and phenanthrene-equivalent PAH metabolites in coho bile relative to the treatment without plants whereas levels of pyrene and benzo(*a*)pyrene equivalents in coho bile, while higher than controls, were not different among the two bioretention treatments (Fig. 6). The concentration of PAHs in treatment waters was not different among bioretention-treated and control waters (Fig. S1). Metal content of juvenile coho gills was not significantly different ($F(2,8) = 1.439–0.084$, $p = 0.321–0.920$) among controls and fish exposed to runoff treated with bioretention, with or without plants (Table 1), despite large differences in metal concentration among treatment waters (Fig. S2).

5. Discussion

Bioretention is a non-proprietary, relatively inexpensive, and readily transferable green infrastructure approach for treating polluted runoff. Here we have assessed the extent to which soil infiltration prevented adverse ecological impacts in the form of lethal and sublethal toxicity to juvenile coho salmon and their invertebrate prey. For the most part, untreated runoff was highly toxic to all species tested. Conversely, stormwater filtered through bioretention columns was strikingly less harmful to coho, mayflies, and cladocerans. We also found that bioretention treatment reversed reproductive impairment in *C. dubia*, a sensitive sublethal indicator of urban stormwater quality (Ireland et al., 1996; Marsalek et al., 1999; Kayhanian et al., 2008; Corsi et al., 2010; McQueen et al., 2010). We found no additional ameliorating effect of plants on aquatic species survival or reproduction, in part because the soil treatment alone was so effective. Overall, these results demonstrate that bioretention can achieve a central aim of green stormwater infrastructure; namely, preventing harm to aquatic animals.

Highway runoff collected for this study contained a suite of contaminants typical of high-use roads (Shinya et al., 2000), including elevated metals, PAHs, and organic matter (McIntyre et al., 2014). Filtration through the soil columns reduced metals by 30–99%, PAHs to levels at or below detection (>92%), and organic matter by over 40% (McIntyre et al., 2014). Tissue concentrations of metals in the gills of juvenile coho salmon exposed to treated runoff and unexposed controls were not significantly different, suggesting that the metals in the post-treatment exposure waters were biologically unavailable. The bioavailability of metals to aquatic animals is determined to a large extent by the presence of dissolved organic matter which sequesters metal ions (Santore et al., 2001; McIntyre et al., 2008; Linbo et al., 2009). For example, whereas dissolved copper is acutely toxic to peripheral sensory neurons in the lateral line of larval zebrafish (*Danio rerio*) at low dissolved organic carbon concentrations (IC50 = 11.5 ppb copper at 0.1 ppm DOC), it becomes much less toxic with relatively modest increases in DOC (IC50 = 50.3 ppb copper at 4.3 ppm DOC) (Linbo et al., 2009). Untreated highway runoff can have very high levels of DOC (37–400 ppm; McIntyre et al., 2014). Although bioretention treatment reduced metals and DOC, the DOC remaining in the effluent was evidently sufficient to prevent metal accumulation in the juvenile coho gill. Thus metals in these highway runoff samples do not appear to be a significant contributor to toxicity, before (McIntyre et al., 2014) or after soil infiltration.

Storing runoff at 4 °C for 7 d eliminated acute mortality in *C. dubia*. We previously found that this storage period resulted in a very significant reduction in PAHs, likely due to microbial degradation (McIntyre et al., 2014). This suggests that organic contaminants are necessary or perhaps even sufficient to cause the mortality observed from exposure to highway runoff. Organic contaminants (vs. metals alone) were also suspected of causing lethal and sublethal effects observed in zebrafish exposed to untreated highway runoff (McIntyre et al., 2014), and have previously been implicated in studies attempting to identify the putative toxicants in urban runoff (McQueen et al., 2010).

Although asymptomatic, juvenile coho salmon exposed to treated runoff had measurably elevated levels of PAH metabolites in their bile relative to controls. Thus, although PAH levels were at or below detection limits in both the treated and control water samples, a small amount of PAHs likely passed through, or were generated by, the soil columns. The concentrations of PAHs in fish bile are commonly higher than in water; e.g., in past studies comparing PAH levels in the bile of caged fish and co-located passive sampling devices relative to surrounding surface waters (Verweij et al., 2004). Although the levels of PAH metabolites were slightly higher in coho exposed to treated runoff relative to controls, the phenanthrene-equivalents in the bile of these fish were at or below the threshold concentrations for harm in juvenile salmonids (Meador et al., 2008). This reflects the exceptional sensitivity of common biomarkers for diagnosing PAH exposure in fish, including bile chemistry and the upregulation of detoxification pathways involving CYP1a and related metabolic enzymes (Lee and Anderson, 2005).

The toxic impacts of urban runoff on aquatic species are often particularly severe during the first flush of a storm (Marsalek et al., 1999; Kayhanian et al., 2008; McQueen et al., 2010; Mayer et al., 2011). As anticipated from previous studies, the first flush from the storms assessed here killed most or all of the fish and invertebrates. Contaminant concentrations in the first flush may be 20-fold higher than the corresponding event mean concentration (EMC) derived from integrated sampling across an entire runoff event (e.g., Shinya et al., 2000). Although EMCs are a conventional metric in stormwater science, toxicity studies based on EMC values often report little or no biological response (Kayhanian et al., 2008). Future GSI effectiveness studies should focus on peak contaminant concentrations in the first flush to minimize underestimates of baseline toxicity and maximize the likelihood of detecting differences in pre- and post-treatment water quality.

Our present results notwithstanding, several important questions specific to the effectiveness of green stormwater infrastructure remain. In the case of bioretention, for example, these include the relative influence of different soil compositions (Carpenter and Hallam, 2010), the influence of biota (particularly plants and microbes; Endreny et al., 2012; LeFevre et al., 2012; Barrett et al., 2013; Palmer et al., 2013), and performance consistency between laboratory and field installations (Carpenter and Hallam, 2010). Another practical consideration, from the perspective of real-world maintenance, is the performance of bioretention systems on a timescale of month to years. Initial long-term effectiveness studies, while limited in number, indicate that metals removal persists on a timescale of decades (Ingvertsen et al., 2012; Paus et al., 2014). While more work is needed in the above areas, our current findings provide preliminary evidence of the biological effectiveness of GSI technology, using metrics that could be expanded to include additional species and endpoints (e.g., endocrine disruption or behavioral changes in fish). Framing effectiveness in terms of aquatic animal health is a relatively novel approach to validating GSI technologies, and our initial findings suggest considerable promise for the success of these types of mitigation methods.

In closing, this and previous studies highlight the considerable hazard that untreated urban runoff poses for aquatic species in receiving waters. In the urban environment, there are often important constraints on the amount of land available for treating stormwater. As a small footprint and relatively inexpensive mitigation technology, the soil columns used here prevented lethal and sublethal toxicity to juvenile coho salmon and their invertebrate prey in response to runoff from a section of densely used four-lane highway. With appropriate scaling (e.g., the installation of sequential bioretention systems along transportation corridors), it may be possible to considerably reduce the harmful toxic impacts of this type of urban runoff.

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Appendix A. Supplementary material

Supplementary data associated with this article can be found, in the online version, at http://dx.doi.org/10.1016/j.chemosphere.2014.12.052.

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