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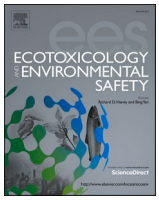

Toxicological effects of building surface runoff on three aquatic species from different trophic levels

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ABSTRACT

Exterior building materials contribute to stormwater runoff pollution but knowledge of how ecological impacts may vary between different types of building materials remains limited. This study combined chemical analyses of runoff from seven different building surface materials with toxicological response analyses as a contribution to addressing this knowledge gap. Results indicate a range of inorganic (e.g. copper, zinc) and organic substances (e.g. diisononyl phthalate, DINP, and nonylphenol) are mobilised by runoff, with concentrations varying between differing materials and rain events by up to three orders of magnitude. Toxicological analysis involving algae, daphnids and fish embryos, indicated that acute and chronic effects also varied between building materials and events, as well as species. For example, copper sheet runoff (maximum concentration 2900 μg/L) exhibited the strongest acute toxic effect on all three test organisms (≥80 % effect irrespective of event and species). Chronic reproductive effects were reported for *Daphnia magna* on exposure to PVC and bitumen felt roof runoff. Results show that runoff from several building surface materials commonly found in urban areas can cause acute and chronic effects on aquatic organisms. Findings could support users to identify environmentally sustainable building materials as a contribution to reducing pollution emissions from cities to receiving waters.

1. Introduction

Stormwater is one of the main transport pathways for pollutants generated in urban areas to enter receiving water bodies, such as lakes and rivers, and exterior building materials are recognised as one of the major contributors of pollutants to stormwater runoff (Müller et al., 2020). Urban areas are typically built using a wide range of materials of varying compositions, functions, and properties. For example, metallic building materials have been documented as one of the largest contributors of metals to runoff since the 1980s (Malmqvist, 1983). Other material types commonly found in urban areas have also been shown to contribute pollutants to runoff, including, e.g., plastic and rubber-based materials such as ethylene propylene diene monomer (EPDM) (Winters et al., 2015) and polyvinyl chloride (PVC), as well as bitumen-based roofing felt or shingles (Müller et al., 2019).

Despite the fact that nature-based solutions (NBS) are recognised as best practice for stormwater management (e.g., in the recast EU Urban Wastewater Treatment Directive of 2024), the direct drainage of stormwater to receiving waters remains the most common approach (Butler et al., 2018). Many of the substances commonly found in building runoff have potential or even confirmed negative effects on aquatic organisms and are therefore listed as priority substances under, e.g., the EU Directive on Priority Substances in Surface Water (2013/39/EU), or similar regulatory guidelines. For instance, several metals commonly found in stormwater are toxic to aquatic organisms (Makepeace et al., 1995; Gillis et al., 2014) and a study of the chemicals most likely to threaten the health of the aquatic environment in the UK found that Al, Cu and Zn posed the greatest risk (Johnson et al., 2017). Moreover, these toxic effects depend on a variety of factors in addition to their concentration, including the bioavailability of the substances (Smith et al., 2015) and the simultaneous effects of several different substances, known as *cocktail effects* (Masoner et al., 2019). However, impacts of actual runoff to freshwater ecosystems have not been thoroughly evaluated from acute or chronic perspectives (Cojoc et al., 2024). Toxicity impacts can be estimated using two approaches: a chemical-specific approach, where chemical water quality is compared to threshold values (e.g., Revitt et al., 2022), and a toxicity-based approach, where toxicity is measured through biological toxicity testing (SIS-CEN/TR 17105:2017). Runoff toxicity studies to-date include an assessment of the toxic effects of road runoff on zebrafish

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(Danio rerio) (McIntyre et al., 2014) and the crustacean *Daphnia magna* (Kayhanian et al., 2008) both of which identified toxic effects. Popick et al. (2022) also assessed the toxicity of stormwater outfall samples through the use of algal growth inhibition tests (*Pseudokirchneriella subcapitata),* reporting that five of the sixteen samples produced a toxic response. However, knowledge of the impacts of runoff from different building materials on aquatic organisms is limited. LaBarre et al. (2017) studied the toxicity of copper roof runoff towards *D. magna* before and after treatment in bioretention planter boxes and biofiltration swales and found that treatment decreased the risk of adverse effects in receiving waters evidenced by both a reduction in Cu concentrations in the runoff and decreases in toxicity to *D. magna*. In an alternative approach, Heisterkamp et al. (2023) investigated the toxicity of construction products through a combination of leaching experiments and biotests, enabling both the identification of substances suspected to cause toxicity (e.g., phenols and isothiazolinones) and the concentrations at which effects occurred. Their study concluded that the combination of toxicity tests and chemical analyses complement each other in characterising the hazard potential of eluates from building surface materials. Expanding this area of knowledge could contribute to forming a basis for source control and mitigation of potentially negative effects in receiving waters caused by substances released from exterior building materials as well as supporting practitioners to select building materials from a receiving water toxicity-reduction perspective.

The overall purpose of this study was to investigate and compare toxicological effects of runoff from different exterior building materials on aquatic organisms. The aim was to generate new knowledge about which of the investigated building materials can cause negative ecological effects as a result of runoff and its transport as a component of stormwater, and to support the choice of environmentally sustainable building materials. In this study, both approaches to assessing toxicity are combined, i.e., comparing chemical water quality with threshold values, and measuring the toxicological response of building surface runoff on aquatic organisms (algae, crustacea, fish embryos) from three different trophic levels.

2. Methods

2.1. Site description and material selection

This study was conducted at an existing experimental setup on the campus of Luleå University of Technology (Sweden). In brief, this consists of pilot roofing panels, each with individual gutters for runoff collection (for full details see Müller et al., 2019, 2023). Runoff from seven different exterior building materials commonly found on roofs and facades in the urban environment (sheets of copper (CUS), zinc (ZNS), galvanised steel (GAL) and stainless steel (STS); bitumen-based roofing felt (BIF); and PVC roofing membrane from two different manufacturers (PVA and PVB)) was collected from three rain events (see Table 1). The selection of these seven materials was based on previous studies of 11 different building materials at the same experimental set-up, where the quality of runoff with respect to the chemical/physical parameters pH, conductivity, particle content, as well as the content of metals, alkylphenols and phthalates, was determined. These studies showed that the main contributors of copper and zinc were CUS, ZNS and GAL, while PVA and PVB contributed phthalates and alkylphenols, and BIF contributed alkylphenols to runoff (Müller et al., 2019, 2023). In these previous studies as well as in the present study, STS was used as a control material to investigate the contribution from atmospheric deposition. Further details about the experimental setup and sampling methods may be found in Müller et al. (2019, 2023).

Table 1
Characteristics of the three sampled rainfall events.

Rain event	Date	Precipitation (mm)	Duration (h)	Antecedent dry days
1	2019–08–30	9.7	7	5
2	2023–06–29	8.9	2	5
3	2023–09–12	3.0	6	8

2.2. Runoff sampling

The total runoff volume generated by each of the seven 2 m^2 individual panel during three rain events (August 2019, June 2023 and September 2023; see Table 1) was collected in Perfluoroalkoxy alkanes (PFA) bags as one single sample per panel, corresponding to the event mean concentration (EMC). Thus, a total of 21 runoff samples were tested within the present study. The collected sample volume varied with precipitation depth, and was approximately 17 L, 16 L, and 7 L for the three rain events, respectively. To achieve sufficient sample volume for the following physicochemical and chemical analyses (see Section 2.3) samples collected from replicate panels of the same materials were combined into one sample for event 3, as previous analyses of physicochemical and chemical runoff quality showed generally low levels of variation between triplicate panels (Müller et al., 2019, 2023).

The samples were collected as soon as possible after the end of each rainfall, but at the latest within 15 h, during which the samples were not exposed to sunlight (nighttime and covered by roof panel). Samples were mixed thoroughly and distributed in bottles (glass or plastic, depending on the analysis). After collection, fresh samples were kept cool (4 ± 1°C) and dark until analyses for chemical quality parameters (see Table 2), while the samples for toxicological testing were kept frozen (-18 ± 1°C) until analysis (see Table 3).

2.3. Physicochemical and chemical analyses

Electrical conductivity (EC), pH, and total suspended solids (TSS) were analysed in the Environmental Laboratory of Luleå University of Technology as soon as possible (within a maximum of 8 h) after sample

Table 2
The analytical methods and reporting limits (RL) for the chemical analyses of the runoff samples.

Analysed parameter (RL, µg/L)	Analytical method
Total suspended solids, TSS (2 mg/L)	Vacuum filtration through 1.6 µm glass fibre filters (SS-EN 872:2005) The filters were oven dried at 105°C ± 2°C and weighed for TSS determination.
Total elements: Al (10), As (0.5), Ba (1), Ca (200), Cd (0.05) Co (0.2), Cr (0.9), Cu (1), Fe (10), K (400), Mg (200), Mn (0.9), Mo (0.5), (Na) (500), Ni (0.6), Pb (0.5), V (0.2), Zn (4)	HNO_3 autoclave digestion; ICP-AES (Ca, K, Mg and Na); ICP-SFMS (other elements), (SS EN ISO 17294–2: 2016, SS-EN ISO 11885:2009 and US EPA 200.7:1994)
Phthalates: dimethylphthalate, diethylphthalate, di-n-propylphthalate, di-n-butylphthalate, diisobutyl phthalate, di-n-pentylphthalate, di-n-octylphthalate, di-(2-ethylhexyl) phthalate, butylbenzylphthalate, dicyclohexylphthalate, diisodecyl phthalate, diisononyl phthalate, and di-n-hexylphthalate (1)	Hexane extraction; GC-MS (standard DIN ISO 18856)
Alkylphenols and -ethoxylates: 4-tert-octylphenol, 4-tert-octylphenol monoethoxylate, 4-tert-octylphenol diethoxylate, 4-tert-octylphenol triethoxylate (0.01), 4-nonylphenols (tech. mixture), 4-nonylphenol monoethoxylate, 4-nonylphenol diethoxylate, 4-nonylphenol triethoxylate (0.1)	Acidification and dichloromethane extraction; GC-MS (based on the ISO standard 18857–2).

Key: ICP-SFMS = inductively coupled plasma-sector field mass spectrometry; ICP-AES = inductively coupled plasma-atomic emission spectrometry; GC-MS = gas chromatography-mass spectrometry (GC-MS).

Table 3
Summary of the toxicological tests and their respective standard method.

Test	Method
Growth inhibition test, green alga *P. subcapitata* (72 h; acute)	SS-EN ISO, (8692): (2012)
Acute toxicity on eggs of zebrafish, *D. rerio* (48 h)	SS-EN ISO (1508)8: (2008)
Acute toxicity, crustacean *D. magna* (48 h)	SS-EN ISO, (6341): (2012)
Reproduction test with the crustacean *D. magna* (21 days; chronic)	EN ISO (1070)6:(2000)

collection. Samples were submitted to an accredited laboratory (ALS Scandinavia AB) for the analysis of metals, alkylphenols and phthalates. The analytical methods are summarised in Table 2.

2.4. Toxicological tests

The toxicological tests were performed by an accredited laboratory (Toxicon AB). Assays evaluated effects on three aquatic organisms, algae, crustacea and fish, from different trophic levels, in order to develop a clear view of the toxic effects following the approach recommended by CEN/TR (1710)5:(2017) 'Guidance on the use of ecotoxicity tests applied to construction products'. The tests performed on the 21 runoff samples, with the standard methods for each test are summarised in Table 3 and described in greater detail in Section 2.4.1; 2.4.2; 2.4.3; and 2.4.4. All acute toxicity tests were performed using an initial limit test (i.e., using the highest possible sample concentration). The *D. magna* reproduction tests (indicating chronic effects) were performed only on samples that did not show any acute toxic effect.

2.4.1. Growth inhibition of Pseudokirchneriella subcapitata

The tests were carried out in accordance with standard SS-EN ISO, (8692):(2012), '*Fresh water algal growth inhibition test with unicellular green algae*'. The limit tests were at a sample concentration of 90 % by volume. Six replicates were used for each sample as well as for laboratory controls (nutrient medium and algae only). Microscopic cell counts were performed on all samples at 24, 48, and 72 h after the start of the test, as a measure of cell density and to calculate growth rates. A reduced growth rate of 5 % or higher, relative to the laboratory control, was considered a growth inhibitory effect as specified by SS-EN ISO, (8692–2012).

2.4.2. Acute toxicity on eggs of zebrafish Danio rerio

The tests were carried out in accordance with standard SS-EN ISO (1508)8: (2008) '*Determination of acute toxicity of wastewater to zebrafish eggs (Danio rerio)*'. Freshly fertilised eggs (embryos) of zebrafish, *D. rerio*, were used in the tests, which were performed as limit tests, where the maximum sample concentration was 100 % by volume. Negative controls (reverse osmosis water (<10 mS/cm) with added salts: 294 mg/L $CaCl_2$ x 2 H_2O; 64.8 mg/L $NaHCO_3$; 123 mg/L $MgSO_4$ x 7 H_2O; and 5.8 mg/L KCl) and positive controls (3.7 mg/L 3,4-dichloroaniline) were performed together with the other samples. Ten replicates of one egg per replicate was performed for each sample including controls. After 48 h, the number of dead embryos was observed. An individual was considered dead if the egg had coagulated, if tail detachment was not reached after 48 h, or if no pulse was observed. The test samples are considered to have a toxic effect on zebrafish eggs at a mortality rate of 10 % or higher.

2.4.3. Inhibition of mobility of the crustacean Daphnia magna

The tests were performed in accordance with standard SS-EN ISO, (6341):(2012). '*Determination of the inhibition of the mobility of Daphnia magna Straus (Cladocera, Crustacea) - Acute toxicity test*'. The method involves incubating newly hatched animals, 6–24 h old, for 48 h and observing mobility in the highest possible test concentration of the sample (limit test; 100 % sample), or - if the proportion of immobilised crustaceans exceeds 10 % in the limit test - a concentration series of the sample (full-scale test) is recommended. The full-scale dilution series includes testing mobility inhibition of six sample concentrations: 3.13; 6.25; 12.5; 25; 50 and 100 % v/v. Four replicates with five animals in each were used for the control and for each sample concentration. Mobility inhibition is determined after 24 and 48 h. (EN ISO 6341). The EC_{50} values, i.e. the concentration at which 50 % of the crustaceans are immobilised, were determined using graphical interpolation to provide point estimates only. The lowest ineffective dilution (LID), corresponding to the highest concentration tested at which no more than 10 % of the crustaceans are immobilised, was also determined.

2.4.4. Chronic toxicity to the crustacean Daphnia magna

The tests were carried out in accordance with standard EN ISO 10706 '*Determination of long term toxicity of substances to Daphnia magna Straus (Cladocera, Crustacea)*' on the samples that did not show acute toxicity to the same species, meaning that survival was sufficient for chronic limit tests to be performed. In the chronic toxicity tests, young females (6–24 h at the start of the test and not first-generation offspring) were exposed to the highest possible test concentration of the sample (90 % by volume). The test exposure time was 21 days. Ten replicates of one test animal per replicate were used for each sample as well as laboratory controls. The control and test solutions were renewed three times per week, when the number of live offspring were counted and dead animals removed. At the end of the test, the total number of live offspring produced by each test animal still alive at the end of the test was counted. Offspring produced by test animals that died during the test and offspring that died were not included in the data processing. The reproductive performance of the test animals exposed to the test samples was compared to that of the laboratory control animals to determine if there was a reproductive inhibitory effect in the test samples.

3. Results and discussion

3.1. Water quality

Among the analysed chemical parameters, Cu, Zn, 4-nonylphenols (NPs) and diisononyl phthalate (DINP) were reported at concentrations clearly deviating from those reported in runoff from both the control material (STS) and other tested materials, indicating their releases from the actual material itself: (see Table 4, together with basic water quality parameters pH, EC, and TSS). Earlier studies also showed concentrations of Cu in CUS runoff as well as Zn in GAL and ZNS runoff were significantly elevated compared to the control material (STS) as well as the other materials (Müller et al., 2023). The Cu and Zn data from the three events sampled within the present study follow the same pattern, with the exception of the Cu concentrations from STS in Event 2 (52 μg/L) which was higher than in runoff from all other materials except CUS. Moreover, Cu and Zn analysed in the panel runoff from all materials was mainly in the dissolved (<0.45 μm) fraction (see Supplementary material). Other elements, analysed in the panel runoff, for which results can be found in the Supplementary material, did not show strong deviations from STS. Among the phthalates and alkylphenols analysed, DINP and NPs were the only two substances reported in concentrations exceeding the analytical reporting limits, with one exception: di-iso-decyl phthalate (DIDP) was reported as 50 μg/L in runoff from PVB at Event 1. In runoff from the control material (STS), neither phthalates nor alkylphenols were detected in concentrations exceeding the analytical reporting limits, which is another indication that these chemical parameters were not derived from the surroundings, but released from the actual materials.

3.2. Limit tests of acute toxicity on the three test organisms

Table 5 shows results from the limit tests on the three test organisms

Table 4
pH, electrical conductivity (EC), total suspended solids (TSS) and total concentrations of Cu, Zn, 4-nonylphenols (NPs) and diisononyl phthalate (DINP) in the runoff from the three sampled events. The tests were carried out on total runoff samples. NPs and DINP were only analysed in runoff from selected materials based on results from previous sampling campaigns.

Rain event		Roofing panel material						
		STS	PVA	PVB	BIF	CUS	GAL	ZNS
pH	1	5.2	5.8	5.5	5.8	5.5	5.5	5.5
	2	7.1	6.1	6.5	6.8	5.9	6.3	6.6
	3	6.3	5.8	5.1	5.1	5.3	6.1	5.5
EC (µS/cm)	1	7.3	8.6	6.7	8.6	8.4	10	13
	2	11	9.2	6.2	8.5	8.0	9.4	9.6
	3	8.5	15	8.8	15	15	18	22
TSS (mg/L)	1	2.2	2.6	< 2	5.8	5.2	4.1	2.1
	2	10	23	76	270	23	27	28
	3	36	35	19	32	24	23	53
Cu (µg/L)	1	3.7	3.4	< 1	1.1	1400	1.1	< 1
	2	52	6.8	1.7	4.8	1900	2.9	3.4
	3	2.5	2.7	1.7	2.1	2900	3.6	3.0
Zn (µg/L)	1	8.6	12	9.1	< 4	10	1800	2700
	2	10	20	12	18	17	1900	2300
	3	11	13	11	8.4	18	3400	5400
NPs (µg/L)	1	< 0.100	< 0.100	1.3	< 0.200	*Not analysed*		
	2	< 0.100	< 0.100	< 0.140	< 0.440			
	3	< 0.100	< 0.120	< 0.300	< 0.370			
DINP (µg/L)	1	< 10	170	200	*Not analysed*			
	2	< 10	31	84				
	3	< 10	130	160				

Table 5
Results from the limit tests on *P. subcapitata, D. rerio,* and *D. magna* as % of affected test organisms. The tests were carried out on total runoff samples. Assessment of which of the samples that were considered to have a toxic effect on the test organisms (in bold) was done in accordance with the respective standard methods (>5 % for P. subcapitata, and >10 % for *D. rerio* and *D. magna)*. '+' indicates stimulatory growth.

Test	Event	STS	PVA	PVB	BIF	CUS	GAL	ZNS
***P. subcapitata* % growth inhibition at 90 % v/v (72 hrs)**	1	+	0.0	+	1.1	**95**	**95**	**97**
	2	**13**	**18**	+	0.72	**89**	**100**	**100**
	3	0.38	+	+	+	**98**	**98**	**95**
***D. rerio* % dead at 100 % v/v sample (48 hrs)**	1	**20**	**20**	**30**	**20**	**90**	10	**30**
	2	**20**	10	**40**	**20**	**100**	**20**	0
	3	0	0	10	10	**80**	10	10
***D. magna* % immobilised at 100 % v/v sample (48 hrs)**	1	**85**	**45**	**100**	**50**	**100**	**100**	**100**
	2	**100**	5	0	0	**100**	**100**	**100**
	3	**100**	**100**	**100**	**70**	**100**	**100**	**100**

as the proportion of affected organisms: % of immobilised *D. magna* after 48 h of exposure; % growth inhibition of *P. subcapitata* after 72 h of exposure compared to the laboratory control, and; proportion of dead *D. rerio* embryos after 48 h. Furthermore, Table 5 shows an assessment of whether the samples were considered to have an acute toxic effect on the respective test organisms (indicated by numbers in bold), i.e., if the growth inhibition of *P. subcapitata* 5% or higher; if more than 10 % of the *D. magna* were immobilised after 48 h; and a mortality rate of 10 % or higher for *D. rerio* embryos, in accordance with the respective standard methods.

From the *P. subcapitata* results presented in Table 5, runoff from all three events for three materials (CUS, ZNS and GAL) consistently displayed a growth inhibition of 89 % or higher: indicating runoff from these materials exerts a toxic effect on *P. subcapitata.* Growth inhibition was below 5 % for four samples (STS Event 3; PVA Event 1; and BIF Events 1 and 2) and therefore not considered to have any growth inhibitory effect on green algae, with runoff from six samples having a stimulatory effect: STS (Event 1), PVA (Event 3), PVB (all three events), BIF (Event 3). Two samples: STS and PVA (both from Event 2) had a growth inhibitory effect of 13–18 %. In all tests, growth was exponential in the laboratory controls (algal mass increased on average 87–258 orders of magnitude for the six controls, see Table A2 of the Supplementary material). Figures A1-A6 of the Supplementary material show photos of all samples at the end of the *P. subcapitata* growth inhibition tests. For instance, Figure A4 shows the control and four of the samples from Event 2 at the end of the test, where there is a clear growth of green algae in the laboratory control (left) and the BIF runoff sample (next to the control), while the other three samples to the right (CUS, ZNS and GAL) show no visible growth of green algae. The biomass and growth rates for the *P. subcapitata* in all samples, including controls, are reported in Table A2 of the supplementary material.

Whilst the *P. subcapitata* results for CUS, GAL and ZNS are approximately comparable per material between events, the response to STS, PVA, PVB and BIF was more variable; e.g., from growth inhibition to a stimulatory effect suggesting the composition of the runoff varies more between events for these materials than for CUS, GAL and ZNS. One important factor for the toxic response of *P. subcapitata* from STS runoff from Event 2 may be the > 1 magnitude higher levels of copper compared to the other two events. It is also noted that PVA and PVB (the same material type but from different manufacturers) did not perform consistently between events, with one sample having a growth inhibitory effect, a further contributing factor being the potential for variations in material composition.

Regarding *D. rerio*, all eggs in the laboratory negative control groups from all three analysed events were alive after 48 h, and for the laboratory positive control group (exposed to 3.7 mg/L 3,4-dichloroaniline), 80, 60 and 50 % of the eggs died for the three analysed events, respectively. The CUS runoff samples from all three events, led to 90, 100 and 80 % of dead embryos after 48 h (see Table 5), respectively, which was a higher impact than that reported for the positive control

group and runoff from all other materials suggesting that, across the substances reported, Cu at the test concentration levels is particularly toxic to *D. rerio* eggs. In contrast, whilst the comparatively high Zn concentrations in runoff from both roofing materials GAL and ZNS seemed to impact *P. subcapitata* growth negatively (Table 5), *D. rerio* eggs, seem less sensitive to high Zn concentrations. Aside from CUS, runoff from PVB from the first two events and ZNS from Event 1 showed the highest toxic effect on *D. rerio*, ranging between 30 % and 40 %. Studies have also reported that impacts of metals can vary between species (Hurle et al., 2006) and even life stages of the same species (Meland et al., 2013), and similar variations in metal susceptibility could be responsible for the effects observed here.

For *D. magna,* the limit tests (100 % by volume) were performed on all 21 runoff samples, with 18 of the 21 samples assessed to have an acute toxic effect ranging from 45 % to 100 % (Table 5), indicating that across all species and material types, *D. magna* is the most sensitive species to a range of building material derived runoff. The 18 samples with an acute response were then subjected to a full-scale dilution series (six sample concentrations), with chronic toxicity tests performed on the three samples (Event 2 runoff from PVA, PVB and BIF) that did not display a toxic response in the limit test, see following Sections 3.3 and 3.4. In these limit tests, laboratory controls for *D. magna* displayed 0–5 % mobility inhibition.

3.3. Mobility inhibition of Daphnia magna – full dilution series

Full-scale dilution series (six concentrations) for mobility inhibition on D. magna were performed on the 18 samples identified as acutely toxic by the limit tests (Table 5). Lowest ineffective dilution (LID) and EC_{50} values after 48 h incubation of the samples are presented in Table 6. Lowest LID and EC_{50} values, indicating the relatively highest toxicity, were displayed for CUS runoff from all three events.

Based on the results for EC_{50} after 48 h (Table 6), the acute toxicity of the runoff from the different materials to *D. magna* was assessed. Following criteria developed by the Swedish Environmental Protection Agency (EPA), EC_{50} values of > 100 were considered to correspond to negligible toxicity, 70–100 low toxicity, 20–70 moderate toxicity, and < 20 high toxicity (Swedish Environmental Protection Agency (EPA) (EPA), 2010). Based on these criteria, toxicity towards *D. magna* is categorised as follows: runoff samples from STS, PVA, and BIF (all three from event 1) had negligible acute toxicity; STS (events 2 and 3), PVA (event 3), PVB (event 3), BIF (event 3) demonstrated low toxicity; PVB (event 1), ZNS (events 1, 2, and 3), GAL (events 1, 2, and 3) and BIF (events 1, 2, and 3) had moderate toxicity; and CUS (events 1, 2, and 3) had high toxicity. Thus, the CUS runoff samples from all three events were assessed as highly toxic to *D. magna*. The runoff from ZNS and GAL from all three events, as well as PVB, Event 1, was assessed to be moderately toxic, while all other samples were assessed to have negligible or low toxicity according to the above categorisation, indicating that the toxicity to *D. magna* varies both with material type and between events. These findings again support earlier findings which indicate that Cu and Zn concentrations are responsible for toxic impacts on aquatic organisms (e.g., Hurle et al., 2006; Crabtree et al., 2009; Johnson et al., 2017).

3.4. Chronic toxicity to Daphnia magna

For three of the samples (PVA, PVB, and BIF, all from Event 2), no acute toxic effect was shown in the limit test on *D. magna* (Table 5). Therefore, the reproduction tests (which are chronic exposure tests) was performed on these three samples only at the highest possible sample concentration. Chronic effects on the reproduction of *D. magna* were observed when exposed to samples of runoff from PVA, PVB, and BIF. Several replicates of each of the three samples had dead offspring, which were not included in the count of the number of offspring as death is considered an acute impact. Moreover, offspring of a parent animal that dies during the test were not included in the calculation. These results are presented in Fig. 1, which show that the sample with runoff from PVA had the greatest negative chronic impact on the number of live offspring per live parent compared to the laboratory control, followed by PVB and BIF. The reduction in the number of offspring was significantly different from the control in all three samples (Anova, $p < 0.05$. Post hoc: Tukey HSD/Kramer).

The results presented in Fig. 1 indicated a chronic response to *D. magna* from all three samples, and highest toxicity for the PVA and PVB samples. These results, combined with the chemical quality of the samples (Table 4) suggest that the presence of DINP may have contributed to a chronic toxic response to *D. magna.* DINP is an endocrine disruptor, and reproductive and developmental effects caused by DINP have previously been found on other species, including *D. rerio* (Tsai et al., 2023; Godoi et al., 2021). Particles from flexible polyvinylchloride, containing DINP have also been reported to affect *D. magna* growth and number of offspring (Schrank et al., 2019). Moreover, long-term exposure of *D. magna* to the other phthalates di (2-ethylhexyl) phthalate (DEHP), dibutyl phthalate (DBP) and diethyl

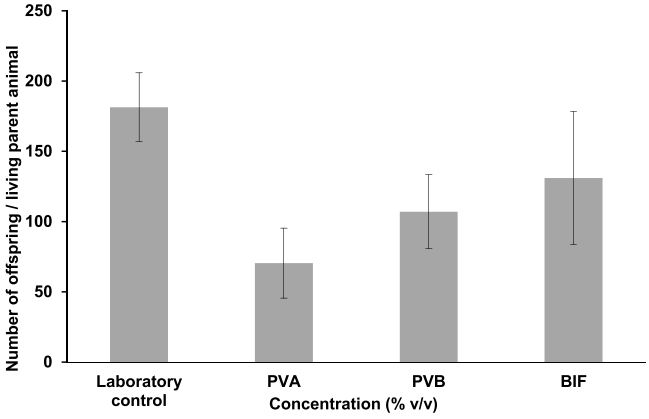

Fig. 1. Mean number of live offspring per live parent in the laboratory control and the three samples PVA, PVB and BIF, all from Event 2. Error bars represent the standard deviation (10 replicates). The tests were carried out on total runoff samples.

Table 6
Lowest ineffective dilution (LID) and EC_{50} values (% v/v) for the tested samples after 48 h of exposure. The LID corresponds to the highest concentration tested at which no more than 10 % of *D. magna* are immobilised. Three samples from Event 2 are missing, as these were not considered to have an acute toxic effect and were instead tested for chronic toxicity (see Section 3.4). Numbers in bold correspond to high toxicity according to Swedish EPA criteria. The tests were carried out on total runoff samples.

	Event	STS	PVA	PVB	BIF	CUS	GAL	ZNS
LID (% v/v)	1	3.1	6.3	50	50	< 3.1	25	25
	2	50	*Not analysed*			3.1	25	25
	3	50	50	50	50	3.1	6.3	13
EC_{50} (% v/v)	1	> 100	> 100	69	> 100	**4.4**	53	59
	2	71	*Not analysed*			**7.6**	36	61
	3	71	100	74	79	**4.3**	50	20

phthalate (DEP) also resulted in negative effects of growth, reproduction and lifespan (Seyoum & Pradhan, 2019).

3.5. Environmental effects

Runoff from CUS was found to have the relatively strongest acute toxic effects on all three test organisms, followed by ZNS and GAL, which had acute toxic effects on two out of three test organisms: *P. subcapitata* and *D. magna* at all three sampled events, as well as for one event each for *D. rerio*. LaBarre et al. (2017) tested the acute toxicity of runoff from a copper roof on *D. magna*, showing 100 % mortality after 48 h of testing. However, the same study showed that passing the runoff through stormwater control measures (e.g., swales) significantly increased the survival (LaBarre et al., 2017). McIntyre et al. (2014) tested the acute toxicity of stormwater runoff from a highway (AADT = 94 000) in the USA, where concentrations of copper and zinc ranged between 23 and 300 μg/L dissolved Cu and 60–2200 μg/L dissolved Zn, respectively), and found high mortality in zebrafish embryos: up to 100 % in two out of six events, as well as other sublethal effects (e.g., reduced growth and delayed hatch) for all the six events analysed. In the present study, the Cu concentrations from CUS and Zn concentrations from ZNS and GAL (Table 4) were of comparable level to those reported by McIntyre et al. (2014), or even higher, and the proportion of dissolved (>0.45 μm) Cu and Zn were 75 % or higher in all runoff samples from CUS, ZNS and GAL. Furthermore, previous investigations of the speciation of Cu and Zn in runoff from the same pilot panels (CUS and ZNS) showed that the dissolved Cu and Zn was primarily present as free ions, i.e., the most bioavailable form (Lindfors et al., 2017). Kayhanian et al. (2008) also studied the toxicity of road stormwater and estimated that about 90 % of the measured toxic effects of road stormwater on the crustacean species *Ceriodaphnia dubia* were caused by the presence of copper and zinc. This is in keeping with other studies which have suggested that Cu and Zn are responsible for the majority of toxicity in road runoff (e.g., Hurle et al., 2006; Crabtree et al., 2009). Thus, even though it is very challenging to assign toxic effects to specific substances (Heisterkamp et al., 2023), these results support the growing literature which indicates that Cu and Zn exerted the toxic effects on the test organisms.

STS provided a control material to investigate toxic effects of runoff from a material that was expected to primarily contribute pollutants derived from atmospheric deposition and had in previous studies shown low levels of pollutants in the runoff compared to the other materials studied (Müller et al., 2019, 2023). In this study, STS runoff generally showed comparably low toxicity to the three test organisms, although STS runoff was assessed to have an acute toxic effect on *P. subcapitata* in Event 2, *D. rerio* in Events 1 and 2, and all three events for *D. magna* (limit tests, Table 5), but low toxicity to *D. magna* in all three events according to the Swedish EPA criteria for EC_{50} values (Table 6). The elevated Cu concentrations in STS runoff from Event 2 (see Table 4), for which potential explanations include atmospheric deposition or splashing from adjacent panels because of high rain intensity, may explain the toxic response to *P. subcapitata,* since Cu is known to cause growth inhibition of algae such as *P. subcapitata* (e.g., Machado et al., 2015). Moreover, LaBarre et al. (2017) suggested that low water hardness could cause toxic effects in *D. magna*, which could be one of the reasons also in this study (water hardness was estimated based on dissolved Mg and Ca concentrations, and was calculated to between 1 and 5 mg/L for all samples, i.e., classified as soft water (Boyd; 2015)). However, aside from the Cu concentrations at Event 2 as well as the concentrations with clear indication of releases from the tested materials (Cu from CUS; Zn from ZNS and GAL; DINP from PVA and PVB), the water quality data for STS did not stand out from those of the other materials tested (see Table 4 and Table A1 of the Supplementary material). Thus, STS can be considered a control material in terms of chemical runoff quality, but even those materials with a relatively low contribution of the analysed substances may exert comparatively low toxic responses to the test organisms due to other factors, e.g., synergistic effects.

The chemical water quality presented in Table 4 was compared to toxicity thresholds from the NORMAN network database, and water quality thresholds from various organisations reported in the NORMAN database, together with the EU Water Framework Directive environmental quality standards (EQS). The data presented therein supports the notion that the substances present in elevated concentrations in the runoff samples exerted the observed toxicological effects on the test organisms. For instance, the Cu concentrations in CUS runoff (1400–2900 μg/L; see Table 4) is more than three orders of magnitude higher than the predicted no-effect concentration (PNEC) for Cu in freshwater (1 μg/L). The corresponding PNECs for 4-NP (0.043 μg/L) and DINP (0.0067 μg/L) are well below the concentrations found in PVA and PVB runoff. However, it should be noted that these PNECs are also below the reporting limits for the analyses. The DINP no observed effect concentration (NOEC) for *D. magna* (freshwater chronic) reported in the NORMAN database was 34 μg/L, which is in the same magnitude, or lower than the DINP concentrations in PVA and PVB runoff (31–200 μg/L; see Table 4), thus, indicating that the DINP concentrations contributed to the chronic toxic responses observed for PVA and PVB runoff.

The results presented in this study show that several of the tested building surface materials may potentially cause adverse effects on aquatic organisms in receiving waters. Dilution effects and changes in speciation both in the stormwater network and in receiving waters make the actual effects difficult to predict, thus, pointing out a need for future studies including acute and chronic tests of diluted samples of building surface runoff. For instance, LID results presented in Table 6 indicated that copper surface runoff would have to be diluted to < 3 % for less than 10 % of *D. magna* to be immobilised. Future studies should also include development of more detailed concentration-effect data to enable the use of e.g. Probit analysis to calculate EC_{50} values with confidence intervals (e.g. Kang et al., 2022). Moreover, the environmental fate and impact of building surface runoff also depend on the quality status and ecological sensitivity of each individual water body. Treatment measures close to the source are well placed to reduce the risk of negative effects. For instance, McIntyre et al. (2014) showed that the toxic effects on zebrafish embryos from road stormwater were drastically reduced after the stormwater was treated in a biofilter. Similarly, LaBarre et al. (2017) investigated bioretention as a mean of reducing toxicity of copper roof runoff and found that while the mortality of *D. magna* neonates from the inlet runoff samples was 100 %, the corresponding mortality was around 10 % in samples from the treated outlet water.

4. Conclusions

Toxic effects of the collected runoff samples were observed on all three test organisms included in the study. Runoff from the copper sheet was found to have the relatively strongest acute toxic effect on all three test organisms, followed by zinc sheets and galvanised steel, which had toxic effects on two out of three test organisms: *Pseudokirchneriella subcapitata* and *Daphnia magna*. The acute toxic effects were primarily attributed to the high concentrations of copper and zinc reaching up to 2900 μg/L Cu in copper sheet runoff, and 5400 μg/L Zn in zinc sheet runoff, respectively. PVC roofing runoff and bitumen felt roofing runoff both showed chronic effects on the reproduction of *Daphnia magna,* expressed as reproductive inhibitory effect compared to the laboratory control, of which the greatest effect was seen for one of the PVC materials: PVA. Even those materials with a relatively low contribution of the analysed substances, e.g., the control material stainless steel, may exert comparatively low toxic responses to the test organisms due to other factors. These results show that runoff from several building surface materials commonly found in urban areas may potentially cause adverse effects, both acute and chronic, on aquatic organisms where untreated runoff is discharged to receiving water bodies. The results presented in

this study could support the choice of environmentally sustainable building materials and could also be integrated into stormwater quality models to consider impacts in receiving waters through, for example, incorporation of dilution effects to enable potential ecological effects to be predicted. Future studies including testing chronic toxicity on diluted samples of building surface runoff would help estimating actual long-term adverse effects of discharge of untreated building runoff to surface water bodies.

CRediT authorship contribution statement

Heléne Österlund: Writing – review & editing, Methodology. **Lian Lundy:** Writing – review & editing. **Maria Viklander:** Writing – review & editing, Methodology, Funding acquisition. **Alexandra Müller:** Writing – review & editing, Writing – original draft, Methodology, Investigation, Funding acquisition, Conceptualization.

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Declaration of Competing Interest

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Appendix A. Supporting information

Supplementary data associated with this article can be found in the online version at doi:10.1016/j.ecoenv.2025.119610.

Data availability

Data will be made available on request.

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