# Loading, transport, and treatment of emerging chemical and biological contaminants of concern in stormwater

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## ABSTRACT

Stormwater is a largely uncontrolled source of pollution in rural and urban environments across the United States. Concern regarding the growing diversity and abundance of pollutants in stormwater, as well as their impacts on water quality, has grown significantly over the past several decades. In addition to conventional contaminants like nutrients and heavy metals, stormwater is a well-documented source of many contaminants of emerging concern, which can be toxic to both aquatic and terrestrial organisms and remain a barrier to maintaining high quality water resources. Chemical pollutants like pharmaceuticals and personal care products, industrial pollutants such as per- and polyfluoroalkyl substances, and tire wear particles in stormwater are of great concern due to their toxic, genotoxic, mutagenic and carcinogenic properties. Emerging microbial contaminants such as pathogens and antibiotic resistance genes also represent significant threats to environmental water quality and human health. Knowledge regarding the transport, behavior, and the remediation capacity of these pollutants in runoff is key for addressing these pollutants *in situ* and minimizing ecosystem perturbations. To this end, this review paper will analyze current understanding of these contaminants in stormwater runoff in terms of their transport, behavior, and bioremediation potential.

**Key words** | antibiotic resistance genes, emerging contaminants, pollutant transport, stormwater, tire wear particles

# HIGHLIGHTS

- Emerging chemical and biological contaminants threaten environmental and human health.
- PFAS pollution in stormwater is growing.
- Tire wear particles have been putatively identified as contaminants responsible for pre-spawn mortality in salmon.
- Stormwater may be acting as a reservoir of antibiotic resistance genes.
- Enhancing the bioremediation capacity of green stormwater infrastructure is needed.

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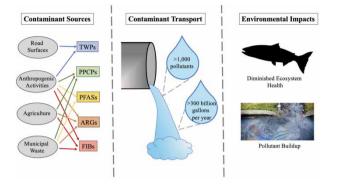
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#### **GRAPHICAL ABSTRACT**



# INTRODUCTION

Over the past several decades, increasing urbanization has dramatically amplified the footprint of impermeable surfaces, generating large volumes of highly polluted stormwater runoff and leading to significant alterations in the natural hydrologic cycle as well as several instances of poor surface and groundwater quality (Bai et al. 2018). Stormwater runoff remains a significant driver of environmental pollution and is a major source of several new contaminants of emerging concern (CECs) such as pharmaceuticals and personal care products (PPCP), tire wear particle (TWP) compounds, and per- and polyfluorinated alkyl substances (PFASs). These CECs are not addressed under conventional water quality regulations and their source and loading patterns vary greatly. Despite the dangers they pose to humans and the environment, little to no information is available regarding their distribution, abundance, and possible eco-toxicological health effects in urban stormwater, though the body of literature around these contaminants is growing. The type of pollutants within stormwater is largely influenced by environment type (i.e. urban vs. rural) and soil chemistry (National Research Council 2009), which drives infiltration and sequestration of stormwater contaminants across large geospatial scales (Mikkelsen et al. 1997). The degree of stormwater pollution is also highly correlated with key factors of urbanization such as geographic location, traffic volume, and land use. Despite past research efforts into managing stormwater pollution, stormwater management practices remain an imperfect solution to a growing problem (Liu et al. 2017). This is partially a result of the narrow pollutant focus of many studies, which has primarily been limited to removing legacy organic contaminants (e.g. polycyclic aromatic hydrocarbons (PAHs), polychlorinated biphenyls (PCBs)), metals, nutrients, and fecal indicator bacteria and ignores many common emerging chemical and biological contaminants of interest. However, as a result of urbanization and industrialization, stormwater is being contaminated by increasingly complex chemical mixtures, which are continually released into runoff from a wide variety of point sources (Fairbairn et al. 2018). These trends are particularly pronounced in urban watersheds, where infiltration and groundwater recharge can be minimal. For example, more than 370 billion gallons of stormwater containing more than 5,000 unique pollutants enter Puget Sound in Washington State annually (Peter et al. 2018; Tian et al. 2020) whose behavior and transport remain poorly understood in the environment (Fairbairn et al. 2018). Many of these micropollutants also pose long-term health risks, even at dilute concentrations (Tran et al. 2019). Similarly, while there are numerous studies on the chemical contaminants of stormwater, there are far fewer studies related to biological contaminants of emerging concern (BCECs) (i.e. antibiotic resistance genes, pathogens) or stormwater-associated microbial communities and their bioremediation potential. Current projections estimate that up to 90% of the global population could be living in urban centers before the end of the century, indicating that observed trends for stormwater pollution are expected to get worse, posing substantial potential risks to environmental and human health (National Research Council 2009). Understanding the sources, pathways, and consequences of stormwater pollutants is critical for designing stormwater management systems that adequately address issues related to stormwater quality and quantity (Liu et al. 2017). This review paper will summarize the transport, behavior, and remediation potential of selected emerging contaminants, and biological contaminants in urban runoff, as well as the potential impacts on environmental and human health.

#### **CEC transport and loading in stormwater**

Emerging contaminants make their way to stormwater through diverse point sources including residential lawns and gardens, animal wastes, industrial activities, and transportation, among others (Tran *et al.* 2019) (Figure 1). Runoff also typically follows a 'first flush' pattern, during which time stormwater becomes highly contaminated with CECs within the first hour of rainfall (Zushi & Masunaga 2009; Tran *et al.* 2019). The abundance of these pollutants was highly correlated to seasonal changes in anthropogenic activities and hydrology; pesticides and PPCPs were most frequently detected in summer months while recalcitrant pollutants (e.g. PAHs, halogenated compounds) were more frequently detected in winter months (Zushi & Masunaga 2009). Release patterns for individual CEC classes are discussed below.

#### Per- and polyfluorinated compounds

Perfluoroalkyl acids (PFAAs) and per- and polyfluoroalkyl substances (PFASs) are synthetic organofluorine compounds containing C-C bonds and C-F bonds. These compounds also possess both a hydrophobic carbon-fluorine tail and a polar and hydrophilic non-fluorinated head (Siegemund et al. 2000), giving them ambivalent properties and making environmental transport and behavior modeling difficult. PFASs and PFAAs have been widely used across the globe for diverse applications such as for the production of nonstick cookware, components of firefighting foam, stain-resistant carpets and fabrics, coatings for packaging, industrial applications, and other personal products since the 1940s because of their strong chemical stability and amphiphilic properties, which simultaneously repel water and oil (Schultz et al. 2003). PFASs are frequently released into the environment during the manufacture and

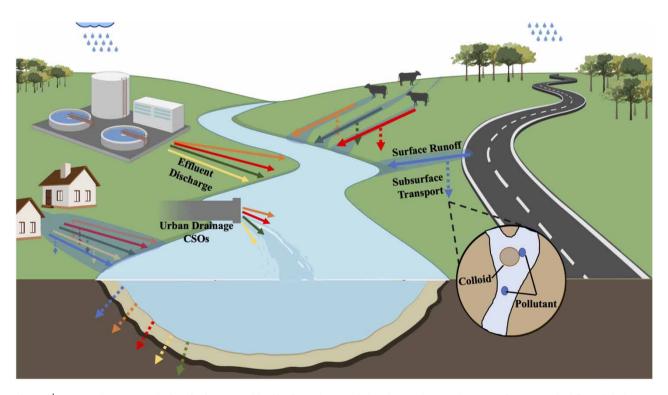


Figure 1 Sources and transport mechanisms for tire wear particles (blue lines), pharmaceuticals and personal care products (green lines), per- and polyfluoroalkyl substances (yellow lines), antibiotic resistance genes (orange lines), and fecal microbes (red lines) in stormwater. Solid lines represent potential direct (e.g. contaminant collection in surface runoff) or indirect (e.g. contaminant release in wastewater effluents or leaking drainage pipes) transport routes to stormwater and receiving waters. Dashed lines represent potential routes of subsurface transport to soils and groundwater via infiltration. The full color version of this figure is available in the online version of this paper, at http://dx.doi.org/10. 2166/wst.2021.187.

anthropogenic use of primary and secondary PFAS products (Agency for Toxic Substances and Disease Registry 2018). As a result, PFASs and their precursors have been detected in nearly every aquatic environment including urban runoff (Kim & Kannan 2007), groundwater (Schultz et al. 2003), wastewater (Sinclair & Kannan 2006) as well as biota, mammals contained within them (Giesy & Kannan 2002; Austin et al. 2003; Hölzer et al. 2008), highlighting the interconnectivity and complexity of transport pathways (Nguyen et al. 2011). Within the United States, the most heavily contaminated sites are located in the northeast region due to the historic use of PFASs and GenX in Minneapolis, Ohio, and Virginia (Nguyen et al. 2011). PFASs have recently been implicated in a variety of health problems within humans and animals, making them priority CECs for future stormwater management and treatment technologies. Internal exposures to PFASs has been linked to several health impacts such as decreased sperm count, low birth weight, endocrine disruption, thyroid disease, certain cancers in both humans and animal models (Austin et al. 2003; Post et al. 2012; Agency for Toxic Substances and Disease Registry 2018). Recent literature indicates these compounds are highly persistent in the environment as well as resistant to abiotic or biological transformation due to the enormous amount of energy required to break C-F bonds (i.e. >110 kcal/mol) (Siegemund *et al.* 2000). This low potential for natural physicochemical and biological treatment in the environment also suggests they are capable of being transported, inert, across vast distances once captured within stormwater runoff.

PFASs and PFAAs often enter surface and stormwater through a more limited number of point sources such as industrial and municipal wastewater, landfill leachate, and select consumer products (Simcik & Dorweiler 2005; Zushi & Masunaga 2009; Dong et al. 2020). PFAS compounds and their precursors are also widely used as firefighting foaming in airports and military bases (Schultz et al. 2003), providing an additional route of transport that remains challenging to control. Transport to stormwater is thought to occur through two primary mechanisms: (i) release from various point sources (e.g. wastewater treatment plants, surface deposits); and (ii) volatilization into the atmosphere and subsequent capture and deposition in water droplets during rain events (Xiao et al. 2012). Several compounds, including perfluorinated sulfonic acids (PFSAs) (e.g. perfluorooctanesulfonic acid (PFOS, C<sub>8</sub>F<sub>17</sub>SO<sub>3</sub>H)) as well as perfluorinated carboxylic acids (PFCAs) (e.g. perfluorooctanoic acid (PFOA, C<sub>7</sub>F<sub>15</sub>COOH)), have been frequently detected in both urban and rural stormwater at a wide range of concentrations (Nguyen et al. 2011; Houtz & Sedlak 2012) (Table 1). Houtz & Sedlak (2012) investigated urban runoff samples from San Francisco, California, and detected PFOS at concentrations ranging from 2.6 - 26 ng/L. PFOA at concentrations 2.1 - 16 ng/L, and PFHxA at concentrations of 0.9 - 9.7 ng/L. These concentrations are consistent with those detected by Xiao et al. (2012) in Minneapolis-St. Paul, Minnesota, with median PFOA concentrations of 9.3 ng/L with a maximum up to 30.6 ng/L and median PFOS concentrations of 10.6 ng/L with a maximum up to 42.5 ng/L in stormwater runoff. As PFAA precursors are difficult to measure, the authors also exposed samples to hydroxyl radicals to convert precursor compounds to measurable PFCAs and PFSAs. After oxidation, the concentration of PFCA increased significantly by 69% (i.e. up to 56 ng/L), indicating the maximum potential concentrations of PFASs in stormwaters and surface waters to be much higher than previously estimated.

Stormwater has been shown to be the transport mechanism driving PFAS pollution in the Tsurumi River basin near Tokyo, Japan, where 11-fold more PFAS compounds are detected in stormwater runoff relative to wastewater effluents (Zushi et al. 2008). It is also clear that PFASs and PFAAs frequently make their way into stormwater runoff in both free and particulate-bound states, though release patterns vary significantly. Some PFAS species like perfluorooctanoic acid (PFOA) and perfluorooctanesulfonic acid (PFOS) have higher hydrophobicity (log  $K_{ow} = 5.3$ and 6.3, respectively), indicating they have an elevated tendency to enter stormwater bound to particulate matter (Zhao et al. 2016) and maybe more resistant to physicochemical or biological transformation. This is further supported by the demonstration that PFAA and PFAS loading into surface waters have been closely correlated with shifts in traffic volumes, which generate large amounts of particulate matters through friction, preceding and during rain events (Dong et al. 2020). At 25 °C, water solubility of PFOS and PFOA is estimated to be 680 mg/L and 9,500 mg/L, respectively. While these solubility limits are expected to be lower in stormwater and surface water due to the presence of other CECs and particulates, this represents a significant potential reservoir and long-range transport pathway for fluorinated compounds in the environment. This is supported by observed discharge fluxes of PFASs and PFAAs in highly urban environments such as Nanjing City, China and Minneapolis-St. Paul, MN where  $\Sigma$ PFAAs can reach up to 916.5 g/day (Dong *et al.* 2020) and 7.9 kg/year (Xiao et al. 2012), respectively, following runoff events.

#### Table 1 | Classification, incidence, and impacts of emerging contaminants

Class	Example species	Chemical formula	Observed concentration in stormwater	Health impacts	Source	Citation
Tire wear particles	Hexa methoxymethyl melamine (HMMM)	$C_{15}H_{30}N_6O_6$	8 – 13 ng/L during baseflow conditions 30–200 ng/L during storm events	Acute coho salmon mortality	Processed rubbers, tires	Peter <i>et al.</i> (2018), Tian <i>et al.</i> (2020), Tian <i>et al.</i> (2021)
	1,3-Diphenyl guanidine (1,3-DPG)	$C_{13}H_{13}N_3$	1800 ng/L in road runoff, 20 ng/L in surface water		Tire additive	
	1,3-dicyclohexylurea	$C_{13}H_{24}N_2O$	300 ng/L in road runoff, 10 ng/L in surface water		TWP leachates	
	N-methyl- dicyclohexylamine	$C_{13}H_{25}N$	10 ng/L in road runoff, 0.5 ng/L in surface water		TWP leachates	
	1-cyclohexyl-3phenylurea	$C_{13}H_{18}N_2O$	350 ng/L in road runoff, 5 ng/L in surface water		TWP leachates	
	6PPD-quinone	$C_{18}H_{24}N_2$	0.3–19 μg/L in representative roadway runoff and stormwater-affected creeks		Tire rubber antioxidant	
	2-mercaptobenzothiazole	$C_7H_5NS_2$	3.0–170 ng/L		Rubber tire vulcanization	
	benzothiazole-2-sulfonic acid	$C_7H_5NO_3S_2$	47-820 ng/L		Vehicle related	
	benzotriazole	$C_6H_5N_3$	2.2–6.3 ng/L		Corrosion inhibitor	
	5-methyl-benzotriazole	$C_7H_7N_3$	0.4–27 ng/L		De-icing	
PPCPs	Methyl-1H-benzotriazole	$C_{14}H_{14}N_6$	Median conc. 806 ng/L Maximum conc. 5550 ng/L	Endocrine disruption	Corrosion inhibitor	Fairbairn <i>et al</i> . (2018)
	Cotinine	$C_{10}H_{12}N_2O$	Median conc. 54 ng/L Maximum conc. 540 ng/L		Tobacco products	
	N,N-Diethyl-m-toluamide (DEET)	$C_{12}H_{17}NO$	Median conc. 120 ng/L Maximum conc. 490 ng/L		Insect repellent	
	Nicotine	$C_{10}H_{14}N_2$	Median conc. 205 ng/L Maximum conc. 3890 ng/L		Tobacco products	
	Caffeine	$C_8 H_{10} N_4 O_2$	Median conc. 207 ng/L Maximum conc. 1710 ng/L		Nerve stimulator	
	Lidocaine	$C_{14}H_{22}N_2O$	Median conc. 3.94 ng/L Maximum conc. 19.9 ng/L		Anesthetic	
	Acetaminophen	$C_8H_9NO_2$	Median conc. 23.9 ng/L Maximum conc. 2110 ng/L		Analgesic	
	Metformin	$C_4H_{11}N_5$	Median conc. 14.9 ng/L Maximum conc. 247 ng/L		Diabetes medication	
	5-Methyl-1H-benzotriazole	$C_7H_7N_3$	Median conc. 135 ng/L Maximum conc. 1340 ng/L		Aircraft deicing and antifreeze	
PFASs	PFOS	C <sub>8</sub> HF <sub>17</sub> O <sub>3</sub> S	38.7-44.1 ng/L	Decreased sperm count, low birth weight, endocrine disruption	Fire-fighting foams,	Zushi et al. (2008),
	PFOA	$C_8HF_{15}O_2$	22.5–28.4 ng/L		non-stick	Zushi & Masunaga
	PFHxA	$C_6HF_{11}O_2$	0.9 – 9.7 ng/L		coatings	(2009), Xiao <i>et al.</i> (2012)
	PFHpA	$C_7HF_{13}O_2$	5.7–6.4 ng/L		-	
	PFNA	$C_9HF_{17}O_2$	84.9-92.8 ng/L			
	PFDA	$\mathrm{C_{10}HF_{19}O_2}$	1.5–5 ng/L			
	PFUnDA	$C_{11}HF_{21}O_2$	3.3–6.7 ng/L			

Uncertainty remains regarding the influence of transportation activity on the types (e.g. short chain vs. long chain) or state (e.g. adsorbed vs. dissolved) of PFASs and PFAAs that are released into stormwater. Some studies have noted that short chain compounds like perfluorohexane sulfonate (PFHxS) are the dominant form detected in stormwater following heavy traffic events (Dong *et al.* 2020), though additional studies should be conducted to verify these trends. However, in general, short chain PFASs tend to be much less adsorbed in sediments and suspended particulate matter and prevail mostly in water compared to long chain PFASs, and the associated functional group also plays a significant role in this spatial distribution (Zhao *et al.* 2016). Long chain compounds have a higher potential to be adsorbed in soil particles because compound polarity and solubility decrease with increasing chain length and the length of C-F bonds also influences the scavenging rates through wet deposition and consequent long distance travel (Taniyasu et al. 2013). Despite their unique chemical partitioning properties, fluorinated CECs also tend to follow first flush patterns in urban environments across the globe, resulting in most pollutants being mobilized within the first hour of a rain event (Zushi & Masunaga 2009). For example, the Hayabuchi River in Japan receives a high concentration of long-chain-length PFCAs during the first-flush after the rain event. Concentrations of long chain PFCAs such as perfluorodecanoic acid (PFDA) increased up to 3.4-fold higher in the Hayabuchi River during and after runoff events, while short and medium chain PFCAs were detected at similar concentrations throughout runoff events (Zushi & Masunaga 2009). This suggests that the chain length and transport state (i.e. free vs. particulate bound) of these compounds are important factors that determine overall transport behavior or stormwater loading concentrations. A study conducted around Minneapolis-St. Paul, MN, observed similar trends, with a variety PFASs detected in all stormwater samples collected from commercial and industrial locations. PFAS concentration spikes were elevated near industrial areas, with the highest concentrations among PFASs observed for PFOS, for both dissolved (156 ng/L) and particle-bound (590 ng PFOS/g total particulate matter) fractions. Findings were also suggestive of disparate transport pathways across residential, commercial, and industrial settings; atmospheric deposition was determined to be the prime mechanism of PFAS release into residential stormwater, while non-atmospheric deposition (e.g. point source release, surface deposits) drives PFAS release in commercial and industrial settings (Xiao et al. 2012). Notably, rural areas adjacent to industrial facilities that use PFASs are disproportionately affected by PFASs contamination according to a study conducted by Lu et al. (2017). The study reported that PFAS concentrations were higher in groundwater and rainwater compared to surface water, indicating lower photochemical and oxidative degradation of PFAS compounds in a closed environment like groundwater compare to an open system like surface water. PFAS compounds were identified in groundwater, soil, and adjacent river water several kilometers away from the nearest point source, demonstrating its transport potential and the growing need for large-scale research efforts to assess PFAS transport behavior across landscapes, particularly for rural communities (Lu et al. 2017). However, our understanding of the mechanisms underlying the presence, persistence, transport, and accumulation of PFASs from precursor compounds in stormwater runoff is still in its infancy. Further research is needed to understand the pathway and behavior of PFASs in runoff along with the effect of PFASs in the aquatic environment and on human health.

### Tire wear particles

Growing concern over the ubiquity and potential environmental impacts of tire wear particles (TWPs), particulate debris produced from the friction between tires and the road surface, has resulted in significant efforts to understanding their environmental fate and behavior over the past decade. TWPs are categorized as microplastics (i.e. length <5 mm) and contain a mixture of rubber tire tread and material from the road surface, which may contain dozens of additional contaminants released from car activity such as toxic tire additives (Peter et al. 2018). Traffic volumes have risen steadily in urban areas, leading to the widespread generation and release of tire wear particles into the environment. The two most significant dispersal pathways of tire wear particles in the environment are currently understood to be transportation by air and runoff. During typical tire tear and wear, small particles (0.1-10% of total TWPs emission) are typically emitted into the air, while large particles (>10  $\mu$ m), which make up the majority of TWPs, deposit onto road surfaces (Wagner et al. 2018). The deposited TWPs will become trapped in the asphalt pavement or transported by stormwater runoff into soils, sewers, and surface waters, where they can continually leach their chemical constituents into the surrounding environment (Kole et al. 2017). Because nonpoint source pollution from road surfaces often flows directly into surface waters untreated, assessing the fate, persistence, and transport mechanisms are key to understanding TWP impacts on both human and environmental health. TWP microplastics are chemically complex materials containing rubbers, reinforcing agents, resins, additives, process oils, net vulcanization agents, and other compounds that can leach into the surrounding terrestrial or aquatic environment, with poorly understood toxicokinetic and toxicodynamic properties (Wagner *et al.* 2018). Thus, the consequences of TWPs loading in the environment remain complex and difficult to predict or prevent.

Recently, TWPs have been consistently shown to be more pervasive in the environment than previously estimated. Gray *et al.* (2018), demonstrated that 90% of all microplastics detected in the Charleston Harbor (South Carolina, USA) were TWPs leached from nearby highways. An additional study conducted by Sutton *et al.* (2019) in collaboration with the San Francisco Estuary Institute and the 5 Gyres Institute in California found that stormwater is responsible for transporting in excess of seven trillion pieces of microplastics into San Francisco Bay each year, with the majority of these being tire wire particles. Stormwater is currently thought to be the largest driver of TWPs transport in the environment, conveying a microplastic load 300-fold greater than the estimated annual discharge from all wastewater treatment plants discharging into San Francisco Bay (Sutton *et al.* 2019). This is largely due to the fact the vast majority of TWPs are generated on road surfaces and remain largely immobile before being picked up in stormwater runoff.

Tire wear particles (TWPs) contain multiple toxic additives and have been implicated as a potential causative agent of pre-spawn mortality in coho salmon across the Pacific Northwest, indicating that a more robust understanding of stormwater pollutants is critically needed (Peter et al. 2018; Tian et al. 2021). Acute pre-spawn mortality has affected coho salmon in the Pacific Northwest for several decades, with exposure to TWPs in stormwater runoff occurring primarily through ventilation and feeding activities (Kole et al. 2017). In highly urbanized watersheds, prespawn mortality can affect up to 90% of juvenile coho (Feist *et al.* 2017). TWP leachates containing methoxymethyl melamines (e.g. Hexamethoxymethyl melamine (HMMM)) and bicyclic amines (e.g. 1,3-diphenyl guanidine (1,3-DPG)) were identified within the salmon mortality signature using high resolution mass spectrometry. HMMM and 1,3-DPG are commonly used additives in tire manufacturing, with up to 14% of the initial tire mass attributable to these compounds, accounting for up to 2.8 lbs released into the environment per tire (Unice et al. 2015) based on an average passenger tire mass of 20 lbs (Lee et al. 2020). Other identified compounds that may be contributing to coho salmon mortality included 1,3-dicyclohexylurea, N-methyl-dicyclohexylamine, and 1-cyclohexyl-3-phenylurea (Peter et al. 2018). The role of TWPs in pre-spawn mortality has been further supported by a recent study by Tian et al. (2021), which putatively identified a previously unknown anti-ozonant compound, 6PPD-quinone ( $C_{18}H_{24}N_2$ ), as the responsible toxicant once converted into a more toxic form  $(C_{18}H_{22}N_2O_2)$  upon contact with ozone. This protoxicant parent compound also appears to be highly stable in the environment, even the following exposure to temperatures up to  $80 \circ C$ .

Wagner *et al.* (2018), estimated that  $1.1 \times 10^6$  tons per annum of TWPs are generated based on total travel distances and traffic volumes. Given that the average car tire contains an up to 10 phr resin content (Burkholder *et al.* 2006) with the majority of resins containing 6PPD, HMMM, DPG, and dozens of other additives identified by Peter *et al.* (2018), this represents a significant and poorly understood reservoir of potentially acutely toxic CECs. Recent literature suggests that microplastics, including TWPs, also affect other sensitive aquatic organisms like grass shrimp, and effects appear dependent on the size and shape of microplastics (Gray & Weinstein 2017). Humans may also be exposed to TWPs, though the human health impacts remain unclear; the primary pathways of exposure are inhalation and intestinal uptake from food intake containing tire wear particles (Kole *et al.* 2017). Several mechanisms for inhalation and intestinal uptake are available for micro and nanosized particles but there are no specific studies regarding human intestinal uptake of tire wear particles, requiring additional research to fully assess the nature and mechanism of this toxicity.

While much work has been done regarding the fate and persistence of airborne TWPs, the remediation potential of these compounds before they enter aquatic environments is still uncertain (Baensch-Baltruschat et al. 2020). To date, the behavior of TWPs has been primarily studied as a component of bulk microplastics in the environment, with few studies being carried out focusing solely on TWPs (Kole et al. 2017). As a result, there is only limited information regarding the total amount of TWPs in stormwater, wastewater treatment plants, and surface waters or their removal efficiency and ultimate fate in the environment. Despite significant advances in our understanding of the role TWPs play in acute salmon toxicity and the putative identification of primary toxicants, installing point source controls or treatment systems along major highways is likely cost-prohibitive and infeasible.

#### Pharmaceuticals and personal care products

PPCPs continue to be significant emerging contaminants in stormwater due to the wide variety of chemical properties observed among PPCP classes as well as their persistence, bioaccumulation, and toxicity potential observed for acute exposures. The per capita usage of PPCPs has steadily increased all across the world over the past several decades, which has ultimately enhanced the discharge of active and inactive pharmaceutical substances into the environment via stormwater (Hopkins & Blaney 2016). The EU Water Framework Directive has identified 33 priority pollutants, including several PPCPs such as iopamidol, ibuprofen, bisphenol A, triclosan, and carbamazepine, as priority CECs to control in stormwater runoff, based on the potential risk to receiving water bodies. Pharmaceutical compounds are frequently the most commonly detected pollutant in stormwater around metropolitan areas (Bai et al. 2018; Tran et al. 2019). Researchers analyzed 31 contaminants of emerging concern from treated and untreated wastewater, urban and agricultural stormwater runoff as well as freshwater bodies and concluded that PPCPs and artificial sweeteners were the most common compounds (Tran et al. 2019). They also found some similar and dissimilar compounds in urban and agricultural runoff due to the varying land use pattern. In addition to being derived from non-point sources, many pharmaceuticals enter stormwater as wastewater treatment plant (WWTP) effluents (Ebele et al. 2017) since conventional WWTPs cannot completely remove PPCPs or they are transported directly from animal excreta to runoff and receiving waters (Boxall et al. 2003; Kaczala & Blum 2016). Overall, PPCPs tend to be more hydrophilic and less volatile than other CECs (Launay et al. 2016; Ebele et al. 2017), making long-range transport possible and even likely. Fairbairn et al. (2018), identified 123 unique CEC compounds in stormwaters sampled in Minneapolis-St. Paul, Minnesota. Of these, 31 CECs were detected in half of the samples with the majority being commercial and consumer care compounds, pharmaceuticals, and pesticides, which typically enter stormwater as a direct result of anthropogenic activities. After initial use, many PPCPs are released into the environment through wastewater treatment plants effluents, combined sewer overflows, and landfill (Hopkins & Blaney 2016). A recent study by Hopkins & Blaney (2016) identified 59 unique PPCPs in WWTPs that were subsequently detected in downstream surface waters, agricultural soils, and landfills following the land application or disposal of wastewater-generated biosolids. As a result, there have been a number of instances of PPCPs captured in stormwater runoff being transported from agricultural soils across long distances or transported directly into groundwater (Kleywegt et al. 2007). One example of that is groundwater contamination with carbamazepine, galaxolide, and bisphenol A in Saale, Germany, by urban stormwater recharge, local river water infiltration and sewer exfiltration (Osenbrück et al. 2007).

The primary concerns with stormwater-driven PPCP loading in the environment are the potential for bioaccumulation and endocrine disruption in humans and animals (Hopkins & Blaney 2016). Many PPCPs such as estradiols, steroids, and bisphenol A are known endocrine disrupting compounds (EDCs) that alter the natural functions of hormones and result in diverse health effects in aquatic and terrestrial organisms (Hayes *et al.* 2002). The presence of EDCs in stormwater has had significant consequences for aquatic ecosystems, as they can significantly alter hormone levels in multiple developing aquatic organisms even at very low concentrations, often resulting in sub-lethal reproductive effects such as decreased fertility, feminization, and hermaphroditism that may not be observed until adulthood. Viglino et al. (2008) observed that estrogenic hormones including natural estrogens, estradiol and estrone, and synthetic estrogen 17-alpha derived from human and animal excreta, have often exceeded the recognized threshold concentrations for the disruption of sexual characteristics of fish, suggesting that retrofitting WWTPs and animal husbandry operations to implement stormwater infrastructure for enhanced point source control measures may have the most positive impact on limiting the spread of EDCs in the environment. Though environmental concentrations are much lower than the consumer application levels, environmental and human health may also be threatened by the hydrophobicity and consequent bioaccumulation potential of many PPCP compounds (Hopkins & Blaney 2016). The two most frequently used chemicals in fragrances - nitro musk and polycyclic musk, frequently detected in stormwater runoff - exhibit Kow values of 4.0-4.4 and 5.4-5.9, respectively. This indicates they have moderate to high risk of bioaccumulation in terrestrial and aquatic species located farther up the food chain. Many emerging PPCPs such as those used in sunscreens, lotions and cosmetics also have log Kow within this range, with many studies suggesting they should be priority pollutant targets when optimizing future stormwater infrastructure.

These trends were also observed in a study conducted by de Solla et al. (2016), which positively identified more than 40 PPCPs from multiple pharmaceutical classes (e.g. stimulants, antibiotics, and antibacterial agents) in wild and caged mussels' tissues both upstream and downstream of a WWTP near Grand River, Ontario, further supporting that WWTP point sources as well as nonpoint sources contribute to PPCP loading in the environment. They observed bioaccumulation factors ranging from 0.66 for diabetes medications (i.e. metformin) to 32,022 for certain antidepressants (i.e. sertraline). The effect of these and similar selective serotonin reuptake inhibitors (SSRIs) in aquatic organisms showed the LC<sub>50</sub> value is 0.14 mg/L for sertraline in aquatic microcrustacean Ceriodaphnia dubia. The same experiment also revealed that sertraline had the lowest observed effect concentration (i.e. 0.045 mg/L), which decreased the number of offspring produced, compared to other four serotonins (citalopram, fluoxetine, fluvoxamine, and paroxetine). Increased risk of intersex characteristics has also been observed in fathead minnow populations, likely due to exposures to high concentrations of metformin (Niemuth et al. 2014). The widespread disposal of antibiotics into landfills and greywater has also raised concerns and has led to a marked increase in the number and abundance of antibiotics documented in stormwater over the past several decades, likely contributing to the spread of environmental bacterial resistance to these antimicrobials and antibiotics (Garner et al. 2017). The consumption of PPCPs is increasing day by day and recent research studies demonstrate that PPCP transport in stormwater runoff and loading in the environment is also rising, creating an urgent need for stormwater treatment systems that mitigate these compounds (Hopkins & Blaney 2016). At present, most of the studies conducted on PPCPs are related to the role of wastewater treatment plants in releasing PPCPs, and very little information is available regarding the long-range transport of PPCPs in stormwater runoff as well as their ultimate fate in the environment. Further research is necessary to understand the pathway and behavior of PPCPs in runoff along with the effect of PPCPs in the aquatic environment and on human health.

Overall, these studies indicate that stormwater may be acting as a significant reservoir of priority CECs like PPCPs, TWPs, and PFASs that remains poorly understood. Rapid urbanization has simultaneously decreased the abundance of pervious surfaces and increased anthropogenic activity in and around urban watersheds. This has served to increase the number and diversity of point sources available to leach CECs into runoff, resulting in the heightened complexity, loading, and transport of pollutants. Over the past several decades, the types of novel and legacy compounds that tend to be transported in stormwater have also shifted due to the limited space available for infiltration, resulting in pollutants being consistently detected far away from their source of origin.

# **BIOLOGICAL CONTAMINANTS**

#### Pathogens and fecal indicator bacteria

In addition to transporting traditional CECs, stormwater can also act as a significant driver of the long-range transport of microbial contaminants like pathogens derived from fecal contamination including *Pseudomonas aeruginosa* and *Staphylococcus aureus* (Table 2) (Parker *et al.* 2010). Pathogens enter stormwater through multiple transport pathways including leaking sanitary sewer lines or septic systems, road runoff, or animal waste (U.S. Environmental Protection Agency (EPA) 1999). High numbers of fecal indicator bacteria (FIB) in urban stormwater runoff can diminish water quality at various receiving waters such as streams, lakes, estuaries, urban beaches and rivers (U.S. Environmental Protection Agency 1999; Noble et al. 2006) and pose a significant human health risk. Following runoff events, FIB levels in receiving waters are often documented to exceed water quality standards across the globe, including the United States, Australia, China and Romania (Parker et al. 2010; Sidhu et al. 2012; He et al. 2019). A study conducted by Parker et al. (2010) showed stormwater frequently surpassed recreational water quality guidelines in coastal North Carolina due to the presence of excess fecal bacteria. During multiple rain events, human fecal contamination was identified in stormwater runoff in the same study due to the existence of Bacteroides Human-Specific markers and fecal *Bacteroides* spp. Sauer *et al.* (2011), sampled 45 stormwater outfalls within metropolitan Milwaukee during the 2006-2009 wet-dry seasons. Of these, 66% of collected outfall samples contained more than 10<sup>5</sup> CFU per 100 ml and sewage sources were identified as the major source of pathogens in those outfalls in terms of human Bacteroides to total Bacteroides ratio (Sauer et al. 2011). FIB have also been detected at concentrations several orders of magnitude higher than water quality limits in coastal Australia, with Enterococcus spp. numbers detected as high as  $1.0 \times 10^4$  CFU/100 mL (Sidhu et al. 2012). Other bacterial (i.e. Campylobacter jejuni, Campylobacter coli, Salmonella enterica, and human-specific HF183 bacteria) and viral (i.e. human adenovirus, human polyomavirus) pathogens, further highlighting the role that leaky sewer lines and CSOs may play in the transport of FIB to runoff. The spread of FIB by stormwater varies widely with the land use pattern, climate, and hydrogeological factors and so on (Rochelle-Newall et al. 2015). Tiefenthaler et al. (2010) observed that recreational sites are particularly sensitive to stormwater driven microbial contamination, as these sites were frequently found to have significantly higher concentrations of FIB following runoff events compared to other types of land use. The highest FIB concentrations at these sites occurred during early stages of stormwater runoff, providing evidence that some biological pollutants follow the first flush pattern often seen for traditional chemical pollutants. The dissemination of FIB in stormwater is also dependent on runoff flow rates in addition to the total suspended solids concentration, as FIB flux is partially driven by washing out contaminants from terrestrial landscapes and mobilizing stream bed sediments

#### Table 2 Emerging biological contaminants in urban stormwater

	Example species	Observed concentration in stormwater	Health impacts or resistance target	Sources	Citation
Fecal coliform bacteria	Total coliform <i>E. coli</i> <i>Enterococcus</i> spp. <i>Salmonella</i> spp. <i>Streptococci</i> spp.	$2.39 \times 10^{6}$ MPN per 100 mL $1.20 \times 10^{5}$ MPN per 100 mL $1.00 \times 10^{6}$ MPN per 100 mL 5.7-30 MPN/10 L $5-56 \times 10^{4}$ No/	Intestinal and diarrheal disease	Sewage systems and animal waste	Parker <i>et al.</i> (2010) Parker <i>et al.</i> (2010) Parker <i>et al.</i> (2010) U.S. Environmental Protection Agency (EPA)
	Fecal indicator bacteria	100 ml 10 <sup>4</sup> CFU per 100 ml			(1999) Sauer <i>et al</i> . (2011)
ARGs	sul1 sul2 tet(O) tet(W) erm(F) dfrE PmrE rosB ermB baeS mdtD mdtF mdtL phoQ acrF ceoB mdtB mexB mexD phoP smeR smeB aadA ANT-Ia OXA-12 bacA arnA	<pre>&gt;10<sup>8</sup> gene copies/ L &gt; 10<sup>7</sup> gene copies/L &gt; 10<sup>7</sup> gene copies/L &gt; 10<sup>7</sup> gene copies/L &gt; 10<sup>9</sup> gene copies/L Unknown Unk</pre>	Sulfonamides Tetracyclines Macrolides Trimethoprim Polymyxin Nalidixic acid Multidrug efflux pumps or multidrug efflux pump modulation Aminoglycoside β-lactams Peptides Polymixins	Clinical, agricultural and animal use	Garner et al. (2017), Zhang et al. (2016), McCarthy et al. (2012)

to different receiving water bodies (Stumpf *et al.* 2010). Climate change and associated increases in extreme wet weather are expected to accelerate transport of FIB to surface waters via stormwater, which will likely disproportionately affect developing countries due to inadequate storm or flooding infrastructure and lead to the use of contaminated water for domestic activities (Hofstra 2011). Rural communities, particularly those in developing nations, may struggle most as rural sources (41%) are more contaminated with fecal indicator bacteria than urban sources (12%) and Africa (53%) and South East Asia (35%) have the highest contamination

rate due to the lower overall economic growth, lack of stormwater infrastructure, inadequate sanitation and hygiene, and lack of awareness of the potential impacts of stormwater pollution (Bain *et al.* 2014).

Thus, fecal indicator bacteria contamination events are often highly correlated with stormwater runoff events. The interaction of FIB with sediments and other organic matters, survival, persistence and transport mechanism of FIB in different climatic condition is still not clearly understood. Because stormwater runoff routinely contains high numbers of FIB and other enteric pathogens, more robust treatment and management of captured stormwater is needed before reaching recreational surface waters or use in non-potable applications.

#### Antibiotic resistance genes

In addition to conventional contaminants (e.g. metals, nutrients and organic pollutants), stormwater has recently been identified as a potential reservoir for the spread of emerging biological contaminants such as multidrug resistant bacteria and antibiotic resistance genes (ARGs) (Garner et al. 2017). Antibiotic resistance has been identified by the World Health Organization as a serious concern for public health (WHO 2014). ARGs have been previously shown to spread as a result of natural and anthropogenic activities (Pruden et al. 2012), increasing the potential for horizontal gene transfer in competent microbes and deepening the environmental resistome. Thus, stormwater runoff has the potential to carry additional diverse and novel ARGs that have been captured from different point and nonpoint sources to contacted surfaces (e.g. soils, sediments) and receiving water bodies. ARGs have been consistently detected in river and surface water sediments at elevated levels compared to surrounding soils (Mao et al. 2013; Amos et al. 2014), presenting uncertainty about the sources and mechanisms underlying these differences. In recent years, urban runoff has been identified as the potential anthropogenic source of ARGs in systems (Chen et al. 2013; Zhang et al. 2016; Garner et al. 2017). While there are many studies focused on the dissemination of pathogenic and fecal indicator bacteria in surface water following runoff events, far fewer studies have investigated the relationships between stormwater and ARG dissemination (Hathaway & Hunt 2010; McCarthy et al. 2012; Zhang et al. 2016). Runoff that carried terrestrial resistant strains caused opportunistic pathogens (with resistance to antibiotics, either natural or achieved) to increase in number after rainfall in three aquatic systems in India (Mohanta & Goel 2014). Five ARGs (i.e. sul1, sul2, tet(O), tet(W), and erm(F)) were found to be significantly higher in concentration by Garner et al. (2017) after three storm events in Stroubles Creek (Blacksburg, Virginia). ARG profiles were found to be hypervariable between runoff events: up to 121 unique ARGs were detected in a single sample but only 24 ARGs were detected in every runoff sample. Of all ARGs, only some were correlated with the presence of fecal indicator bacteria (i.e. tet(O) and tet(W)), with others (i.e. sul1, sul2 and erm(F)) having no known associated vector. However, unlike FIB transport, ARG transport does not appear to follow first-flush patterns, likely because FIB originate from a collection of point sources and ARGs are ubiquitously present across many environments. Together with past research, these findings indicate that stormwater runoff can act as a considerable transport vector for ARGs across unconnected landscapes.

Once transported in stormwater, intracellular and extracellular ARGs can settle into soils and sediments. For extracellular ARGs, which are not contained within living microbial cells, adsorption to particulate matter significantly increases the persistence and stability of these genes in the environment (Mao et al. 2013), indicating the persistence and dynamics of microbial contaminants in stormwater is much more complex than previously thought. Intracellular ARGs, contained within microbial cells, have been detected more frequently in Gram-negative isolates compared to that of Gram-positive isolates, demonstrating the differential contributions of microbes to environmental antibiotic resistance (Zhang et al. 2016). The downstream threat to human health is compounded by the fact that antibiotic-susceptible bacteria can acquire ARGs horizontally from bacteriophage infection (i.e. via transduction), live cells (i.e. via conjugation), or assimilation from extracellular DNA (i.e. via transformation) (Zhang et al. 2016; Garner et al. 2017). The recent discovery of genetically modified crop-derived ARGs (i.e. nptII, bla), originating from the consumption of genetically modified crops and their associated food products, in wastewater treatment sludge and biosolids (Gardner et al. 2018; Gardner et al. 2019) indicates that the diversity and abundance of ARG sources in the environment is larger than previously estimated. As sewage contamination of stormwater occurs frequently, this presents another potential avenue for ARGs to be captured and transported in stormwater. However, no studies to date have detected ARGs derived from genetically modified crops or quantified ARG horizontal gene transfer rates in stormwater, suggesting that additional research is still needed. In addition, further studies are required to identify the potential relationship between FIB contaminants and ARGs.

As stormwater runoff carries sediment, nutrients, microbes, and antimicrobials (e.g. triclosan, antibiotics), it presents an environment with sufficient selective pressures to favor the maintenance and transfer of ARGs within stormwater-associated microbial communities. The presence of heavy metals in stormwater can create a similar selective pressure, as genes that confer resistance to metals and antibiotics are often co-located on the same mobile plasmids (Berg et al. 2005). Antibiotics and antimicrobials used in clinical, agricultural and animal feed end up in streams through feces, urine or WWTPs as they often cannot be fully transformed in humans or water treatment systems. This has been driven by the overuse and misuse of antibiotics and ultimately led to significant increases in the abundance and diversity of antibiotic resistant bacteria in both aquatic and terrestrial environments (Martinez 2008). This can have widespread and long-lasting consequences for combatting environmental antibiotic resistance. Increasing evidence indicates resistance to last-resort antibiotics such as carbapenems, tigecycline, and colistin has emerged among gram-negative bacteria in South Africa (Sekvere 2016). The mcr-1 colistin resistance gene has been detected in environmental samples in China, South Africa, and Brazil (Sekvere 2016). Since it was first detected in China, it has subsequently been detected in E. coli and K. pneumoniae isolates from foods, humans and animals across the globe (Liu et al. 2016). There is also concern regarding the spread of the New Delhi metallo- $\beta$ -lactamase (bla<sub>NDM-1</sub>) and its variants because of its hydrolyzing capability to  $\beta$ lactam antibiotics, including carbapenems. Both genes have been detected in surface and drinking waters, which demonstrates their widespread occurrence in the aquatic environment. It has also been found in a study that mcr-1 and  $bla_{NDM-1}$  have the capability to disseminate from source to tap water via a drinking water supply system, indicating ineffectiveness of an advanced water treatment process to remove both mcr-1 and bla<sub>NDM-1</sub> (Khan et al. 2020). The same study also identified that mcr-1 has the capability to transfer from drinking water to mouse gut microbes through a horizontal gene transfer process (Khan et al. 2020), demonstrating the potential for environmental ARGs to directly impact human health and underscoring the interconnectivity of our water resources. No studies to date have determined the long-range transport behavior, persistence, and transformation of these last-resort ARGs in stormwater.

# Potential for enhanced CEC removal through enhanced biodegradation

CECs in stormwater runoff continue to threaten global water quality and require proper treatment technologies to mitigate their potential impacts. Bioremediation is a lowcost, publicly accepted technique that has been used worldwide with varying degrees of success (Vidali 2001), particularly as a component of biophysicochemical treatment within engineered Green Stormwater Infrastructure (GSI). Bioremediation involves the biological degradation and transformation of metals and organic wastes to less toxic forms by living organisms (e.g. microbes and plants). However, efficient bioremediation processes are a function of many environmental factors such as soil type, pH, temperature, presence of oxygen and nutrients, which are often difficult to fine-tune in field-scale systems (Watts 1998; Vidali 2001). GSI - including vegetated strips, constructed wetlands, bioretention systems, and bioswales - has enormous potential to act as enhanced bioremediation systems for the stormwater pollutants discussed in this review. Each GSI system is composed of several key components (e.g. soil and media mixtures, endogenous microbes, and plants) capable of individually or synergistically removing stormwater pollutants. Though the primary mechanism of pollutant removal is currently thought to be physicochemical processes (e.g. adsorption, precipitation and filtration) to remove contaminants through physical sequestration (Shafique & Kim 2017; Jayakaran et al. 2019), there is also significant and unexplored potential to enhance biological treatment by leveraging endogenous GSI microbes through established (i.e. biostimulation, bioaugmentation) and emerging (i.e. microbiome engineering) technologies. Microbiome engineering, the process of manipulating an existing microbial community to achieve a desired structure or function, is similar in principle to bioremediation but differs in the theoretical level of specificity and efficacy (Foo et al. 2017; Gardner & Gunsch 2020). Our ability to characterize and manipulate environmental microbiomes and their associated metabolic processes has been transformed in recent decades with the development of high throughput -omics approaches. Engineering GSI microbiomes will likely require a thorough understanding of community metabolic and transcriptomic processes, intracellular dynamics, population resilience, and spatiotemporal distribution of active biodegraders in addition to standard site characterization efforts. However, though engineering the human gut microbiome has been pursued aggressively over the past decade and has already led to transformative advances in medicine, these concepts have not been explored or tested in an environmental context to date (Foo *et al.* 2017).

Microbes capable of degrading toxic pollutants such as TWPs, PFASs, and PPCPs have been detected in many environmental matrices including raw stormwater, soils, sediments, and plants (Table 3). Bioswales and bioretention cells in particular can act as excellent opportunities for directed in situ bioremediation and microbiome manipulation, as they offer a platform to simultaneously enhance the transformation of metals (a plant-dominated process) and anthropogenic organics (a microbe-dominated process) (Kang 2014). As the media for bioretention cells is already engineered, further manipulating media to promote enhanced pollutant breakdown can be achieved by strategically incorporating necessary nutrients, electron donors, electron acceptors and even exogenous bacteria capable of transforming target pollutants. Within GSIs, these strategies may be adapted to the unique pollutant profiles of urban stormwater through directed manipulation of a site's environmental conditions and microbes, which can work synergistically to degrade pollutants (Watts 1998). Genetic bioaugmentation of endogenous microbes, which is designed to increase the catabolic capacity of environmental microbes through horizontal gene transfer, can also work in some cases (Ikuma & Gunsch 2012). However, the natural bioremediation capacity of stormwater has been underutilized for persistent organic compound removal from urban runoff, particularly within GSI systems. Because stormwater can move across large geospatial scales before entering surface waters or fully infiltrating soils, the high number of pollutants in urban stormwater may lead to the preferential proliferation of microbes that can tolerate and degrade pollutants in stormwater and stormwater infrastructure like GSI media. Thus, urban stormwater may be acting as an untapped reservoir of specialized biodegraders that can be leveraged to improve the performance and resilience of GSI systems. Use of the adaptive nature of microbes and providing them with carbon and energy source bioremediation of stormwater contaminants is expected. Aerobic bacteria such as Pseudomonas spp., Sphingomonas spp., Alcaligenes spp., Rhodococcus spp., and Mycobacterium spp. can degrade reduced compounds such as PPCPs, hydrocarbons, alkanes, and polyaromatic compounds (Ghosal et al. 2016). However, aerobic bacteria are not always capable of fully mineralizing contaminants, leaving some compounds partially degraded. Anaerobic bacteria such as Dehalococcoides spp. can perform reductive dehalogenation of fully or almost fully oxidized compounds such as TCE, PCE, PCBs, and chloroform (Watts 1998; He et al. 2007). There is also a significant difference between the application of fungal and bacterial biodegraders, indicating these microbes may be best utilized for disparate applications or can be used to synergistically degrade recalcitrant compounds. Bacteria may use the organic pollutant as the source of energy and carbon while fungi usually require co-metabolic conditions in which both an energy and carbon source are available for degradation to occur. Fungi use general enzymes (e.g. oxygenases, laccases, peroxidases) that are partially effective for breaking down a wide variety of CECs, while bacteria use specific enzymes that are very effective at partially or completely mineralizing a small number of CECs (Vidali 2001). Bacterial degradation is often more frequently used due to the low cost, resilience, and fast growth, though anaerobic bacteria are less used compared to aerobic bacteria (Vidali 2001).

# Biodegradation potential of per- and polyfluoroalkyl substances

Past research has suggested that PFASs are highly resistant to microbial degradation in soils, sediments, and surface waters due to their amphiphilic properties and high energy C-F bonds (Schultz et al. 2003). However, recent studies have identified several unique microbes capable of partial or complete PFAS metabolism, transforming our current understanding of the biodegradability of these compounds (Key et al. 1998). Key et al. (1998), examined biodegradability of both PFOS and H-PFOS using Pseudomonas spp. and found that microbes could partially defluorinate H-PFOS under aerobic conditions. Several other studies have also concluded that PFOS and PFOA can be partially biodegraded under sulfur limiting conditions. Meesters & Schröder (2004) found that anionic PFOS and PFOA can be efficiently removed under anaerobic conditions, but no removal was observed under aerobic conditions. Most significantly, a 2019 study conducted by Huang and Jaffe reported up to 60% PFOA and PFOS removal using wetland isolates in 100 days (Huang & Jaffé 2019). Acidimicrobium spp. A6 was identified as the main microbial driver of PFOA and PFAS degradation. A6 is widely known for Feammox metabolism, in which it transfers electrons from ammonium  $(NH_4^+)$ to ferric iron, which works as an electron acceptor and reduces it to ferrous iron, converting  $NH_4^+$  to  $NO_2^-$  for energy production (Clement et al. 2005). Ammonium and iron were used as putative electron donors with and without PFOA and PFOS to culture A6 in the laboratory. Concentrations of the two PFASs over time were measured using liquid chromatography-tandem mass spectrometry. Up to 60% of initial PFOA and PFOS concentrations were removed after 100 days, with inorganic fluorine increasing over this

#### Table 3 Contaminants with a high potential for enhanced bioremediation

CEC Class	Compound	Biodegrading microbe	Distribution in the environment	Source
PFASs	PFOA PFAS	Acidimicrobium spp. A6	Wetland bacteria	Huang & Jaffé (2019)
		Isolate Envi 5 <sup>a</sup> Envi 7 <sup>a</sup>	Contaminated soil	Tseng (2012)
	PFHxS	Pseudonomas spp. PS27 and PDMF10	PFAS contaminated soil and groundwater	Presentato <i>et al.</i> (2020)
PPCPs	Carbamazepine (antiepileptic/ psychiatric drug)	Labrys portucalensis F11	Polluted soil (Portugal)	Bessa <i>et al</i> . (2019)
	Ibuprofen (NSAID)	Nocardia spp. NRRL 5646	Garden soil	Chen & Rosazza (1994)
		Sphingomonas Ibu-2	Sewage treatment plant	Murdoch & Hay (2013)
		Variovorax Ibu-1	Soil, drinking water	Murdoch & Hay (2015)
		Patulibacter spp. 111	Activated sludge	Almeida et al. (2013)
		Bacillus thuringiensis B1(2015b)	Soil (Poland)	Marchlewicz <i>et al.</i> (2017)
	Triclosan (biocide)	Penicillium spp. <sup>a</sup>	Air, soil, damp environments	Taştan <i>et al</i> . (2016)
		Sphingopyxis strain KCY1	Wastewater	Lee et al. (2012)
		Nitromonas europaea	Environments with high ammonia and inorganic salt	Roh <i>et al</i> . (2009)
		Sphingomonas spp. PH-07	Activated sludge	Kim <i>et al</i> . (2011)
		Aspergillus versicolor <sup>a</sup>	Food products and damp environments	Taştan and Donmez (2015)
		Nannochloris spp. <sup>b</sup>	Freshwater and marine water	Bai & Acharya (2016)
		Rhodotorula mucilaginosaª	Soil, water	Taştan <i>et al</i> . (2016)
		Dyella spp. WW1	Activated sludge	Wang <i>et al</i> . (2018)
	Naproxen NSAID	Stenotrophomonas maltophilia KB	Soil, surface water, wastewater	Wojcieszyńska <i>et al.</i> (2014)
		Planococcus spp. S5	Activated sludge	Domaradzka <i>et al.</i> (2015)
		Bacillus thuringiensis B1 2015b	Soil of the chemical factory 'Organika-Azot' in Jaworzno, Poland	Górny <i>et al</i> . (2019)
		Pseudoxanthomonas spp. DIN-3	Biological activated carbon process filter	Lu <i>et al</i> . (2019)
TWP	Rubber detoxification	Recinicium bicolour <sup>a</sup>	Coniferous wood, deciduous trees	Leidig <i>et al</i> . (1999)
		Rhodococcus rhodochrous	Soil and contaminated soil	Haroune <i>et al.</i> (2004)
		Corynebateria spp.	Soil, water, blood, human skin	Bredberg et al.
		Pseudomonas spp.	Soil and water	(2001)
		Escherichia coli	Soil, manure, and water	
	Rubber devulcanization	Thiobacillus spp.	Marine and terrestrial habitats	Bredberg et al.
		Pyrococcus furiosus <sup>c</sup>	Aquatic anaerobic hyperthermophiles (high temperature loving) archaeon	(2001)
	Rubber degradation	Streptomyces spp.	Soil and decaying vegetation	Rose & Steinbüchel
		Xanthomonas spp.	Plant tissues	(2005)
		Micromonospora spp.	Soil and water	
		Thermomonospora spp.	Soil, usually on plant debris	

<sup>a</sup>Fungal species; <sup>b</sup>Algal species; <sup>c</sup>Archaeal species.

Downloaded from http://iwaponline.com/wst/article-pdf/83/12/2863/906112/wst083122863.pdf by guest same time period. The researchers also identified that A6 was passing electrons from ammonium to the PFASs that release fluoride ions instead of transferring electrons to ferric iron (Huang & Jaffé 2019). There is an interesting fact that research and restriction of PFAS products usage are limited to a very narrow range of selective long chain PFASs products. Numerous unregulated PFAS products including short chain PFASs are still being used as alternative to regulated or banned long chain PFASs (Wang et al. 2017). These shorter chain PFASs are generally less toxic but their end products exhibit persistence, which has been not leveraged till date. Presentato et al. (2020) used naturally occurring microorganisms from PFAS contaminated soil and groundwater and PS27 and PDMF10 environmental isolates were successful in accumulating PFHxS within the cells upon an opportune selective pressure. This study also indicates that stormwater contaminated with PFASs might have some microbes that are capable of degrading them, though additional research is needed to verify these claims.

Fungi have not been widely tested for their ability to degrade PFASs, but wood-rot fungal groups are well known for their capability of degrading multiple recalcitrant halogenated compounds (Vidali 2001; Tseng 2012). Several fungal strains isolated from aqueous fire-fighting foamcontaminated sites have been identified as potential PFASs degraders in bioremediation systems (Tseng 2012). Isolate Envi 5 transformed about 20% PFOS within 28 days and Envi 7 transformed about 20% PFOS within 14 days, and only Envi 7 could partially transform about 20% PFOA within 14 days (Tseng 2012). This experiment demonstrates that fungi could have better potential in degrading PFASs compared to bacteria. Overall, this study suggests a possible abundance of stormwater microbes that might have degradation potential, such as the fungal strains from the contaminated site.

# Biodegradation potential of pharmaceuticals and personal care products

PPCPs represent diverse classes of compounds including hormones, antidepressants, lipid regulators, anticonvulsants, antibiotics, and antihypertensives, often complicating bioremediation efforts (Cizmas *et al.* 2015). Many PPCPs have been observed to be transformed by microorganisms in WWTPs and the environment (Helbling *et al.* 2010), with many organisms using PPCPs as a carbon or energy source (Onesios-Barry *et al.* 2014). A study by Bessa *et al.* (2019) reported that 95.4% carbamazepine was transformed by the bacterial strain *Labrys portucalensis* F11 in monosubstrate culture and 100% biotransformation was reported in cometabolism with acetate. Antibiotics are persistent in the environment due to their lipophilic and hydrophobic nature in spite of their short lifetime. Regarding microbial degradation of antibiotics, fungi are most promising followed by bacteria (Nnenna et al. 2011), though only bacteria not susceptible to the antimicrobial properties of target antibiotics would be viable targets for biostimulation or bioaugmentation. Ibuprofen has also been shown to be susceptible to biodegradation via advanced oxidation processes; however, this often results in the creation of more toxic byproducts than ibuprofen itself. To date, Nocardia spp. NRRL 5646 (Chen & Rosazza 1994), Sphingomonas Ibu-2 (Murdoch & Hay 2013), Variovorax Ibu-1(Murdoch & Hay 2015), Patulibacter spp. I11 in the presence of yeast extract and tryptone (Almeida et al. 2012), Bacillus thuringiensis B1(2015b) in both metabolic and co-metabolic scenarios with glucose (Marchlewicz *et al.* 2017) have been capable of degrading ibuprofen. though information regarding the toxicokinetics and toxicodynamics of metabolites produced is limited. Regarding biodegradation of antimicrobial triclosan, bacteria showed better efficacy compared to fungi, but fungal strain Penicil*lium* spp. was identified as the most efficient known microbe capable of metabolizing more than 80% of initial triclosan concentrations (Tastan et al. 2016). Other microbes capable of degrading triclosan include Nitromonas europaea (Roh et al. 2009), Sphingomonas spp. PH-07 (Kim et al. 2011), Sphingopyxis strain KCY1 (Lee et al. 2012), Aspergillus versicolor (Taştan & Dönmez 2015) Nannochloris spp. (Bai & Acharya 2016), Rhodotorula mucilaginosa (Tastan et al. 2016) and Dyella spp. WW1 (Wang et al. 2018). Naproxen is also amenable to increased biodegradation under certain conditions such as cometabolic environment (Wojcieszyńska & Guzik 2020). While bacteria, fungi, and algae have the capability to transform naproxen to a certain extent, only bacteria are known to be capable of cleaving its aromatic rings. Stenotrophomonas maltophilia KB2 (Wojcieszyńska et al. 2014) Planococcus spp. S5 (Domaradzka et al. 2015), Bacillus thuringiensis B1 (Górny et al. 2019), and Pseudoxanthomonas spp. DIN-3 (Lu et al. 2019) are examples of known bacterial strains that can degrade naproxen under metabolic and cometabolic conditions.

While biodegraders may be capable of metabolizing multiple PPCPs from the same chemical class, transformation efficiencies and endpoints can vary significantly with differences in PPCP chemical structure. Thus, predicting pollutant susceptibility to biodegradation will be dependent upon the presence of specific enzymatic reactions during biodegradation related to specific chemical structures. The efficiency of biodegradation can also vary greatly with initial substrate concentration for different PPCPs, particularly if substrates display antimicrobial or inhibitory properties, though no strong correlation has been established between biodegradation rates and initial substrate concentrations to date (Onesios-Barry *et al.* 2014). This indicates that higher contaminant concentration does not certainly assure higher degradation and that there is likely a concentration limit concentration, above which PPCP biodegradation fails or slows.

Pharmaceuticals are designed in such a way that they can exert phenotypic effects even at low doses, which increases the risk to aquatic and terrestrial life when they are released in the environment in either unaltered or transformed chemical structures (Cizmas et al. 2015). Depending on the structural properties and sorption characteristics of the PPCPs, these CECs may be completely mineralized (e.g. valsartan), left unaltered (e.g. diatrizole) or partially transformed to metabolites (e.g. ibuprofen) (Xia et al. 2005; Helbling et al. 2010; Redeker et al. 2014). These transformation products of unknown toxicity may increase the probability of their environmental persistence or the consequences of their interactions with surrounding biota. Further research on PPCPs transported by stormwater should focus on parent CECs, their potential degradation metabolites in various environmental matrices, as well as the toxicodynamic and toxicokinetic properties of metabolites.

#### Biodegradation potential of tire wear particles:

Tire wear materials are composed of complex mixtures of rubber fillers, processing aids, reinforcement agents, and additional compounds that have varying degrees of potential for biodegradation (Wagner et al. 2018). Natural rubber is comprised mostly of poly (cis-1,4isoprene) and tires are made through vulcanization, which is a thermochemical process that initiates chemical reactions between rubber polymers, sulfur, and supplementary chemicals (Baensch-Baltruschat et al. 2020). During vulcanization, mono-, di-, and poly-sulfidic cross linkages are formed, which often inhibits bacterial degradation of rubber (Stevenson et al. 2008) and makes devulcanization a key barrier that must be overcome during tire particle biodegradation. Because rubber is the primary material in tires, it is expected that tire wear particles will undergo photodegradation and biodegradation in the field (Baensch-Baltruschat et al. 2020). Past research regarding biodegradation of tire wear particles has primarily focused on rubber that has been detoxified, devulcanized, and degraded sequentially by multiple microbes (Stevenson et al. 2008).

Detoxified rubber has demonstrated a greater success rate for devulcanization (Bredberg 2002). The white rot fungus Recinicium bicolour and bacterial species Rhodococcus rhodochrous, Corynebateria spp., Pseudomonas spp., and Escherichia coli have all been demonstrated to detoxify vulcanized rubber under multiple environmental conditions. Three of the 15 tested fungal species (i.e. Pleurotus sajor-caju, Trametes versicolor, and Recinicium bicolour) were capable of biodegrading Poly-R478, an aromatic polymeric dye polyvinylamine sulfonate anthrapyridone, in the presence of cryo-ground tire rubber (Leidig et al. 1999; Bredberg 2002). Among them, Recinicium bicolour appeared most efficient at breaking down cryo-ground tire rubber materials. Haroune et al. (2004) showed that the bacterium Rhodococcus rhodochrous can degrade 2-mercaptobenzothiazole (MBT), a toxic additive in rubber, producing the less toxic metabolites benzothiazolvlsulphonate and 2-methlthiobenzothiazole (MTBT). Members of Corynebateria and Pseudomonas genera, as well as Escherichia coli, have been shown to break down MBT (Bredberg et al. 2001). Both sulfur reduction and oxidation are proved to be successful for devulcanization. Thiobacillus spp. and Pyrococcus furiosus are capable of oxidizing disulfide linkages created during vulcanization and devulcanize rubber material, making them excellent candidates for a multistep bioaugmentation treatment system. However, for successful devulcanization and contaminant breakdown, microbial growth-inhibiting compounds such as zinc oxide and zinc salts may need to be removed first.

The complete breakdown of TWPs also requires the breaking of polyisoprene polymers within the rubber structure, likely requiring two different groups of microbes to degrade rubber-based materials; one group can create clear zones on latex agar plates and use latex as the sole source of carbon and energy (e.g. Streptomyces spp., Xanthomonas spp., Micromonospora spp., Thermomonospora spp., and Actinomyces spp.) (Rose & Steinbüchel 2005). The second group of microbes (e.g. Gordonia spp., Corynebacterium spp., Mycobacterium spp., and Nocardia spp.) will need direct contact with polymer to degrade the polyisoprene chains, likely making fungal degraders less feasible options. This group metabolizes poly (cis1, 4-isoprene) faster than the previous group of bacteria. Latex-clearing protein (Lcp) and rubber oxygenase (RoxA) enzymes start poly (cis-1, 4-isoprene) cleavage, followed by subsequent metabolic steps. TWPs appear to be biodegradable, though often to varying degrees of success. Over a period of 16 months, 52 and 36% of the polymer in TWPs was degraded in roadside soil and glass beads, respectively, while tread rubbers showed no significant signs of degradation (Cadle & Williams 1980).

Thus, it can be concluded that sulfur-degrading microorganisms are critical for TWP breakdown and must be present or added to GSI soils. These findings also imply that tread-wear particles degrade at a faster rate than tread-rubber itself, though few field studies have been conducted to further support these trends. Atmospheric oxidation was also a significant driver of TWP breakdown, and may serve to accelerate biodegradation in the field (Cadle & Williams 1980). To date, no studies known to the authors have been conducted to assess TWP degradation in stormwater runoff or surface waters. While biodegradation of TWPs appears promising, further research is necessary to identify microbes or that can biodegrade other toxic additives like 1,3 DPG and HMMM.

Many GSI systems have been demonstrated to be effective at sequestering heavy metals, nutrients, FIB, and anthropogenic pollutants such as PAHs and slowing their transport in the environment (DiBlasi et al. 2009). However, uncertainty remains regarding the nature of this sequestration and whether pollutants are being permanently removed or merely temporarily adsorbed to GSI media. Many GSI such as bioretention systems are designed for small areas and, thus, are ineffective for managing regional stormwater and high flow volumes, which limits their efficacy in controlling CECs and CBECs. The efficacy of bioretention cells and their associated biodegrading microbes largely depends upon the climatic and hydrological conditions of a site. As a relatively new method of treating stormwater runoff, there are still limitations regarding standard GSI design practices to address different types of climate, runoff volume and surrounding characteristics. Another important parameter is the successful bioremediation, which requires growth of selected microbes and plants that largely depends upon the site properties such as soil conditions, pollutant concentrations, homogeneity of soil mixture and so on (Lynch & Moffat 2005). As with all other bioremediation strategies, there is the possibility that undesired microbes may outcompete biodegrading microbes and lead to system failure or slowdown. Further research replicating bench scale research findings in the field is needed to continue removing these barriers to sustainable and effective CEC removal from stormwater.

#### Conclusion and future research directions

Past research has consistently demonstrated that CECs such as TWPs, PPCPs, and PFASs along with CBECs such as FIB, ARGs, and antibiotic-resistant bacteria are ubiquitously detected in stormwater runoff, with largely unknown consequences for environmental and human health. Though we do not have a thorough understanding of their transport and fate in the environment, it is clear that stormwater runoff and nonpoint source pollution can often act as a vector to carry these pollutants across vast, unconnected landscapes. While many legacy pollutants are known to be passively removed from stormwater runoff through physical adsorption in GSI systems, this typically promotes temporary pollutant sequestration rather than permanent pollutant breakdown. Many GSI systems remain black boxes with few insights into the biological and physico-chemical drivers of pollutant breakdown, a problem that has persisted as a direct result of the mindset that GSI, like other passive systems, is a simplistic system lacking real opportunity for directed engineering optimization. Thus, there is a growing need to better understand how GSI components (e.g. media, microbiomes, plants) act in concert to limit the effects of various toxicants to develop long-term, effective stormwater treatment. Gaining a better understanding of naturally occurring microbiomes in stormwater and GSI systems, as well as their role in treatment performance, is a critical first step. To date, no studies known to the authors have attempted to characterize stormwater or GSI microbiomes to evaluate and manipulate the bioremediation capacity of endogenous microbes. Additional research that leverages metabolic synergies between microbes and their environment will be essential for improving GSI performance and protecting the quality of our water resources. GSI success in permanently removing CECs through bioremediation or microbiome engineering efforts will necessitate identifying the biotic and abiotic factors that control biodegrading microbiome assembly, promote microbiome proliferation, and result in microbiome functional failure. Research that explicitly validates engineering designs by providing biological relevance to the engineered outcome will also be critical to determining if the output treatment technology is truly protective of ecological or human health.

## DATA AVAILABILITY STATEMENT

All relevant data are included in the paper or its Supplementary Information.

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