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# Impacts of a herring gull colony on runoff water quality from an urban green roof

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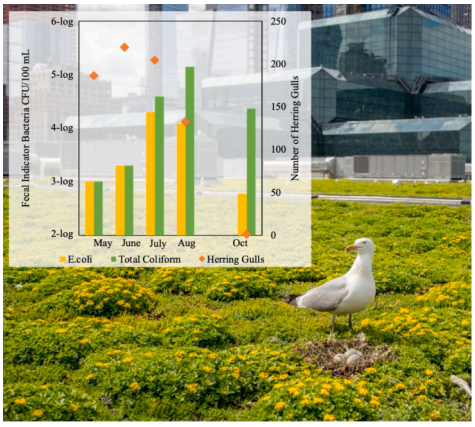
## HIGHLIGHTS

- Javits Center green roof runoff met tested chemical US EPA drinking water standards.
- High fecal contamination exceeded regulatory limits for various water uses.
- Gull presence contributed to fecal contamination on the green roof.
- Runoff from gull-populated green roofs may require treatment before use.

## GRAPHICAL ABSTRACT

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## ABSTRACT

Green infrastructure (GI) strategies, including green roofs, have become a common, decentralized, nature-based strategy for reducing urban runoff and restoring ecosystem services to the urban environment. In this study, we examined the water quality of incident rainfall and runoff from a green roof installed on top of the Jacob K. Javits Convention Center in New York City. Since the 2014 installation of this green roof, one of the largest in North America, a colony of nesting herring gulls grew to approximately 100 nesting pairs in 2018 and 150 nesting pairs in 2019. Water quality monitoring took place between September 2018 and October 2019. Except for phosphorus on some occasions, we found concentrations of nitrate, nitrite, chlorine, sulfate to be below federal drinking water standards. Levels of the fecal indicator bacteria (FIB), total coliform, *E. coli*, and *Enterococcus*, were consistently higher in runoff samples than rainwater, ranging from 150 to over 20,000 CFU/100 mL for *E. coli* and 100 to over 140,000 CFU/100 mL for total coliform. Quantitative polymerase chain reaction (qPCR) methods were used to search for potential opportunistic pathogens, including *Legionella* spp., *Mycobacterium* spp., *Campylobacter* spp., and *Salmonella* spp. Discovery of the presence of *Catellicoccus marimammalium*, a gull-

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associated marker in runoff water indicates that herring gulls are the likely source of contamination. Due to habitat loss, herring gulls, and other *Larus* gull species are increasingly nesting on urban roofs, both green (such as at the Javits Center) and conventional (such as on Rikers and Governors Islands). Habitat creation is one of the target ecosystem services desired from GI systems. Although the discharge from the green roof of the Javits Center is directed to the city's sewer system, this study demonstrates the need to treat runoff from green roofs with nesting gull populations if its intended use involves reuse or human contact.

## 1. Introduction

A form of decentralized green infrastructure (GI), green roofs are a multifunctional, nature-based, stormwater management strategy with a wide range of co-benefits including stormwater retention (U.S. GSA, 2011; Abualfaraj et al., 2018), reduced ambient air and surface temperature (Niachou et al., 2001; Jim and Peng, 2012), thermal buffering (Alvizuri et al., 2017), air pollution improvement (Speak et al., 2012), increased exposure of urban populations to green spaces (Hui, 2006). While most green roofs are designed to detain or retain/reduce stormwater quantities, they are rarely designed to improve the quality of roof runoff.

It is well known that rooftop runoff can be a source of microbiological and chemical contamination (Lye, 2009). Microbiological contamination of water in rooftop harvesting systems, for both conventional and green roofs, can originate from deposition of airborne microorganisms blown in by wind, dust, leaves, fecal material from birds and small animals, as well as dead animals and insects on the roof (Abbasi and Abbasi, 2011). These sources can also contribute chemicals such as nitrogen, phosphorous, trace metals, and particulates (Abbasi and Abbasi, 2011). In urban and industrial centers poor air quality, inorganic and organic contaminants from heavy traffic, industry, incinerators, and smelters can be dry deposited on roofs (Cunliffe, 1998), only to be flushed off the roof as runoff. Roof runoff can also contain other pollutants like metals and nutrients that leach from the roof materials (Clark et al., 2008). Previous research suggests that direct weathering of the roof materials and leaching of accumulated deposits of particulate matter can also be a source of pollution (Uba and Aghogho, 2000; Adeniyi and Olabanji, 2005).

Microbial species found in roof runoff include *Shigella* and *Vibrio* (Uba and Aghogho, 2000), *Salmonella* (Taylor et al., 2000; Uba and Aghogho, 2000), *Campylobacter* (Merritt et al., 1999; Evans et al., 2007), protozoa *Giardia* and *Cryptosporidium* (Albrechtsen, 2002), *Pseudomonas* (Evans et al., 2007), *Legionella* (Simmons et al., 2008), *Leptospira* (Sasaki et al., 1993), and *Aeromonas* (Albrechtsen, 2002). The presence of these organisms suggests that appropriate treatment should be undertaken before rainwater can be declared potable or reused (Uba and Aghogho, 2000).

The quality of rooftop runoff is especially important if it is to be harvested or used, or if there is the chance of human contact. Runoff from conventional roofs can be harvested, stored, and used for a variety of non-potable uses such as cooling of heating, ventilation, and air conditioning (HVAC) systems, landscape and garden irrigation, toilet flushing, and pavement cleaning (Basinger et al., 2010; Monteiro et al., 2016; Rostad et al., 2016). This research focuses primarily on the adequacy of conventional roof runoff volumes to meet certain quantitative needs. Some researchers (Mendez et al., 2011; Razzaghmanesh et al., 2014; Zhang et al., 2014) have studied the quality of green roof discharge, though not with a focus on water quality as a potential constraint to its reuse. In general, these studies suggest that runoff harvested from green roofs could require treatment to improve its water quality depending on its intended use(s). For example, if the harvested water were to be used for irrigation water for urban agriculture, the presence of heavy metals and pathogens in the water would represent human health risks associated through consumption of food crops (Solomon et al., 2002; Amoah et al., 2005; Khan et al., 2013). High salt concentration could also be a concern, as it would reduce water uptake and be toxic to plants (Haering et al., 2009). In addition, HVAC evaporative cooling towers have frequently been identified as breeding grounds of *Legionella pneumophila*, which causes Legionnaire's Disease (Walser et al., 2014), and thus it is important to ensure that water used to supply cooling towers, such as runoff from green roofs, does not contain *L. pneumophila* or other infectious agents.

Green roof water quality may be affected by a variety of design factors including the roof's structural composition, soil substrate and vegetation. It can also be impacted by external factors such as the quality of incident precipitation or applied irrigation and fertilizer (Wang et al., 2017). It is unclear if discharge from green roofs is of superior water quality than runoff from conventional roofs. For example, many researchers have found that green roofs can leach nitrogen and phosphorus (Moran et al., 2005; Hathaway et al., 2008; Van Seters et al., 2009). Carpenter and Kaluvakolanu (2011) compared runoff from asphalt, gravel ballasted, and extensive green roofs and found mean mass values for total nitrate and phosphate from green roof were lower than from asphalt roof. Discharge water quality may be a function of the green roof age and substrate type. New green roofs tend to be a source of pollutants, whereas older green roofs with established vegetation present a lower pollutant load (Rowe, 2011). Established vegetation and substrates can improve the water quality by absorbing and filtering pollutants (Rowe, 2011). Similarly, one previous study found the concentrations of metals from green roof runoff were lower than in urban runoff, but the concentrations of some metals corresponded to moderately polluted natural water (Berndtsson et al., 2006). There is limited research about the microbiological contamination of green roof runoff, but Hussain and Berndtsson (2012) identified intestinal *Enterococci* in the green roof discharge. Moreover, the soil of green infrastructure (GI) may be at risk of exposure to animal feces and chemical contaminants (Joyner et al., 2019) and may finally affect the runoff water quality.

Microbiological contamination of runoff from both conventional and green roofs can result from fecal material produced by animals that use both green and conventional (non-green) roofs as wildlife habitat. Green roofs provide habitat for arthropods (Tonietto et al., 2011; MacIvor and Ksiazek, 2015; Ksiazek-Mikenas et al., 2018; Starry et al., 2018; Dromgold et al., 2020), bats (Pearce and Walters, 2012; Parkins and Clark, 2015; Partridge et al., 2020a), and birds (Eakin et al., 2015; Partridge and Clark, 2018). Arthropods can be abundant and diverse on green roofs, though not as abundant and diverse as at ground level sites (MacIvor and Lundholm, 2011; Braaker et al., 2014; Dromgold et al., 2020). Both birds and bats use green roofs as foraging habitat during migration and during the breeding season (Parkins and Clark, 2015; Partridge and Clark, 2018; Partridge et al., 2020b). While few bird species nest on green roofs (Partridge and Clark, 2018), ground nesting birds do (Baumann, 2006; Baumann and Kasten, 2010; Baumann et al., 2021), including *Larus* gulls, with up to 1700 ring-billed gull nests recorded in a colony on a single 0.74 ha green roof in Chicago (Washburn et al., 2016). Similarly, due to habitat loss, *Larus* gulls nest on conventional roofs (Kubetzki and Garthe, 2007; Soldatini et al., 2008a; Soldatini et al., 2008b; Perlut et al., 2016). *Larus* gulls can be a carrier of *Salmonella* (Butterfield et al., 1983) and *Escherichia coli* (Araújo et al., 2014) and gulls and geese are sources of fecal contamination in beach sand and water (Leeming et al., 1997; Edge and Hill, 2007). The impact of nesting gulls on runoff quality from both green and conventional roofs is currently unknown.

This study examines the water quality of runoff from an extensive (*Sedum* species) green roof installed on the on the Jacob K. Javits Convention Center in New York City. The green roof, completed in the Spring of 2014, is nearly 27,316 $m^2$ (6.75-acre) in size and is capable of retaining several millions of gallons of storm water annually, or 77 % of average rainfall incident to it (Abualfaraj et al., 2018; Jacob K. Javits Convention Center, 2019). Any runoff generated is discharged directly to the municipal combined sewer system. The roof hosts an active herring gull colony from April through September.

To study the water quality of urban green roof runoff and identify the potential pollutant sources, we examined the concentration total carbon (TC), non-purgeable organic carbon (NPOC), as well as anions ($Cl^-$, $NO_2^-$-N, $NO_3^-$-N, $PO_4^{3-}$, and $SO_4^{2-}$) and cations ($Na^+$, $NH_4^+$-N, $K^+$, $Mg^{2+}$, and $Ca^{2+}$). In addition to these chemicals, we also quantified total coliform, *E. coli*, and *Enterococcus* spp. in the samples via culturing methods and used quantitative polymerase chain reaction (qPCR) to quantify the potential opportunistic pathogens via DNA biomarkers. In order to investigate the impact of nesting herring gulls on the water quality of green roof discharge, the gull-associated fecal biomarker targeting the bacterium *Catellicoccus marimammalium* (Lu et al., 2008) was utilized for qPCR tests. Even if no human contact with the runoff or runoff reuse is proposed, this study has important implications for managers of roofs with bird colonies.

## 2. Materials and methods

### 2.1. Site description and sample collection

A chemical and biological analysis was performed on the incident rain on, and runoff discharged from, the Jacob K. Javits Convention Center green roof (Manhattan, New York City, US). The Javits Center has two green roofs, one on the north side and the other on the south side of the building. Precipitation falling on the green roof was collected at three locations on the North green roof using sterilized plastic bins with holes on the side connected to sterile sampling bottles. These sampling devices were located in close proximity to rain gauges installed on the green roof. Runoff leaving the roof also was collected manually by hand using sterile sampling bottles during each rain event at one of the roof drains located on the North green roof (Fig. 1). Collection of drain samples started once the storm began and sufficient flow was observed exiting the outlet. This drain collected water from a portion of the roof that was about 700 $m^2$ and was half covered by green roof and half covered by conventional roof. For each event, at least one rain sample was collected from each rain sampling device and 4–12 drain samples were collected depending on the rainfall of that day. In total, 36 rain samples and 72 drain samples were collected between Sept 2018 and Oct 2019. Samples were stored in 1 L polyethylene bottles and transported on ice to the laboratory immediately after collection (within 6 h) and sample processing commenced immediately upon arrival.

### 2.2. Herring gull surveys

We conducted bird survey transects to monitor herring gull presence on the roof within 2 and 10 days of water quality sampling, as part of regular biodiversity surveys. Transects were conducted by walking across the roof in an east-west direction six times over the extent of the roof so that each bird on the roof could be observed and counted. Transects were conducted from May through October in 2018 and 2019.

### 2.3. Physical-chemical analysis of water quality

Due to the potential of nutrients and organic constituents to leach from the green roof growing medium, the presence of these chemicals in the roof runoff was investigated. In addition, due to the roof's close proximity to the Lincoln Tunnel, the research goals included determining whether organic hydrocarbons could be detected in precipitation falling on the roof.

Water samples were stored in volatile organic analysis autosampler vials at 4 °C until total carbon (TC) and non-purgeable organic carbon (NPOC) analysis was conducted. NPOC and TC were assessed using a Shimadzu TOC(L)-TN. The instrument was operated according to the manufacturer's instructions. The pH and electrical conductivity (EC) were measured via a pH meter and a conductivity meter (Mettler Toledo), respectively. Water samples for anions and cations analysis were filtered through 0.22 μm syringe filters. The concentrations of anions ($Cl^-$, $NO_2^-$-N, $NO_3^-$-N, $PO_4^{3-}$, and $SO_4^{2-}$) and cations ($Na^+$, $NH_4^+$-N, $K^+$, $Mg^{2+}$, and $Ca^{2+}$) were assessed via high performance liquid chromatography (HPLC). A YS-50 Column was used for cation measurement and 4 mM phosphoric acid was used as liquid phase. A SI-52 4E Column and 3.6 mM $Na_2CO_3$ was used for anion measurement. Detailed descriptions of the operation of the TOC(L)-TN and HPLC can be found in (Price et al., 2015; Price et al., 2016).

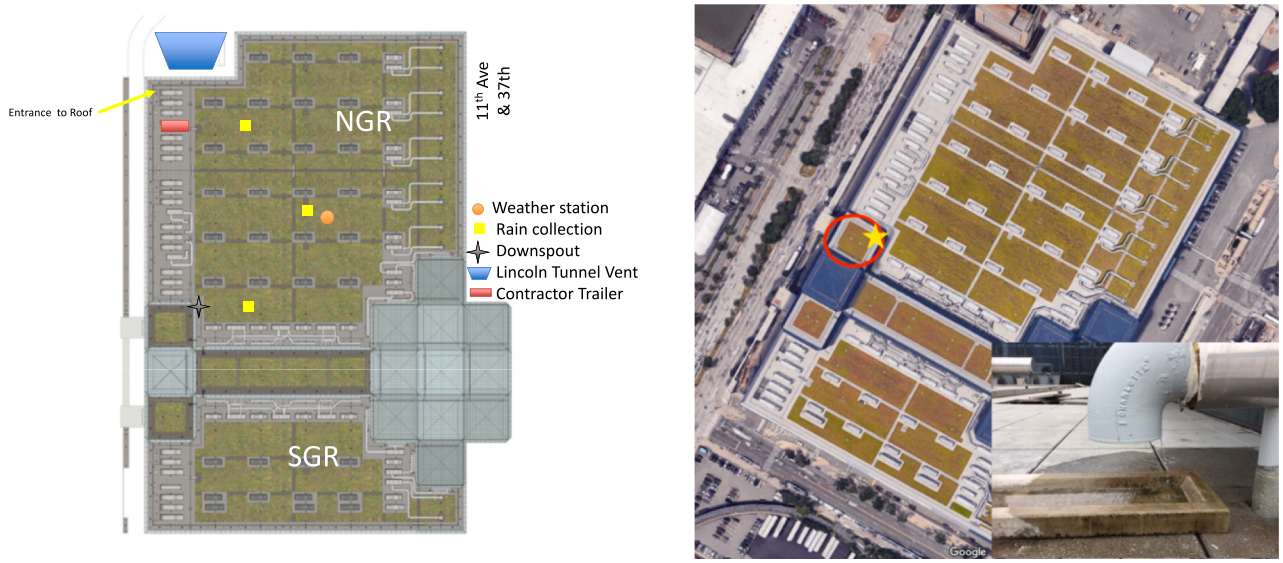


**Fig. 1.** Left is the floor plan of the green of on Jacob K. Javits Convention Center. Right is satellite image of Jacob K. Javits Convention Center. The yellow star denotes the location of the roof drain. The drainage area contributing to this drain is circled in red. Inset is an image of the drain itself.

### 2.4. Bacterial quantification assays

Selective media culturing methods were used to quantify the concentrations of *Enterococcus* spp., total *Coliforms*, and *E. coli*. mEI agar (Difco) and m-ColiBlue (Hach) were used as the culture medium for *Enterococcus* and *Coliforms*, respectively. The 0.22 μm sterile vacuum filters were used to obtain bacteria from water samples. The water sample volume to filter may be different from each rain event (1 mL to 100 mL). The filter papers were put onto the selective agar, then culture overnight at 37 °C.

### 2.5. DNA extraction and quantification

Total DNA was extracted from the water samples to quantify the abundance of *Legionella* spp., *Mycobacterium* spp., *Campylobacter* spp., *Salmonella* spp. and *Catellicoccus marimammalium* via qPCR analyses. Total DNA was extracted by using Qiagen QIAmp Fast DNA Stool Mini Kit following the manufacturer's instructions. The concentration of the extracted DNA for each sample was measured spectrophotometrically via NanoDrop and fluorimetrically via QuBit 2.0.

### 2.6. Real-time PCR (qPCR)

Multiple qPCR primer sets targeting *Legionella* spp. (Leg_F 5′-GCGGCTACCTGGCCTAATAC-3′; Leg_R 5′-CCAACAGTAAGTTGACATCG-3′) (Aoki et al., 2003), *Mycobacterium* spp. (Myco_F 5′-TAGGTCGG-GACGGTGAG-3′; Myco_R 5′-TTGCGAAGTGATTCCTCC-3′) (Tobler et al., 2006), *Campylobacter* spp. (Camp_F 5′-CACGTGCTACAATGGCATATA-CAA-3′; Camp_R 5′-CCGAACTGGGACATATTTTATAGATTT-3′) (de Boer et al., 2015), *Salmonella* spp. (Salm_F 5′-GCTGCGCGCGAACGGCGAAG-3′; Salm_R 5′-TCCCGGCAGAGTTCCCATT-3′) (Ferretti et al., 2001) and *Catellicoccus marimammalium* (Gull_F 5′-TGCATCGACCTAAAGTTTTGAG-3′; Gull_R 5′-GTCAAAGAGCGAGCAGTTACTA-3′) (Lu et al., 2008) (Supplementary Material Table S1) were ordered from IDT DNA and used for qPCR analysis of the DNA extracted water samples. A fast SYBR Green qPCR assay was applied to obtain the concentration of each species. All samples were stored at −20 °C until they were analyzed.

A QuantStudio 3 Real-Time PCR System and Applied Biosystems Fast SYBR Green Master Mix were used to conduct all qPCR assays. Total qPCR reaction volume was 20 μL. Each reaction mixture contained 6 μM of the forward and reverse primers, 2 μL of template DNA and 10 μL of fast SYBR Green Master Mix. The program employed: pre-incubation for 20 s at 95 °C; 40 amplification cycles of 1 s of denaturing at 95 °C and 20 s of annealing at 60 °C; and, finally, 1 s of 95 °C, 20 s of 60 °C and 1 s of 95 °C for melt curve. All assays were conducted in replicates or triplicates of each sample with negative controls (no template) and positive controls.

### 2.7. Statistical analysis

SPSS software package version 26 was applied for all statistical analyses. Independent *t*-test and Welch *t*-test were used to find significant differences between the means of rainwater and runoff water collected on the green roof.

## 3. Results

Climate data for all rain events are presented in Table 1. The results of the water quality analyses performed on rain and runoff samples collected during the 10 rain events from September 2018 to October 2019 are described below.

### 3.1. pH and conductivity

There was no significant difference between the means of pH in rainwater (n = 3) and runoff water (n = 4) (Welch *T*-test, $P = 0.676$) (Supplementary material Table S2), but the rainwater had a larger range of pH. Rainwater displayed an average pH value of 6.96 and runoff displayed an average pH value of 7.4. The minimum and maximum values of pH in the rainwater were 5.21 and 9.28, while the minimum and maximum values of pH in the runoff were 6.35 and 9.

The results showed a significant statistical difference between conductivity of rainwater (n = 8) and runoff water (n = 8) (Independent *T*-test, $P < 0.001$). The runoff had higher conductivity (mean 290.51 mS/cm) than the rainwater (mean 38.95 mS/cm). This finding indicates that salt concentrations were higher in the runoff than in the rain.

**Table 1**
Precipitation, average temperature, total rainfall, and the number of herring gulls present on the Javits Green Roof on each sample collection day.

| Collection date | Days since last precipitation (dry days) | Average temperature [°C] | Total rainfall [mm] |
|---|---|---|---|
| 9/25/2018 | 5 | 15–22.78* | 39.9 |
| 10/11/2018 | 7 | 20.56–25* | 19.2 |
| 11/5/2018 | 2 | 8.89–11.67* | 6.1 |
| 5/13/2019 | <1 | 7.51 | 0.5 |
| 5/28/2019 | 2 | 19.78 | 0.1 |
| 6/18/2019 | 2 | 21.18 | 20.6 |
| 7/17/2019 | 6 | 28.91 | 43.9 |
| 7/23/2019 | 5 | 21.6 | 13.4 |
| 8/7/2019 | 3 | 23.93 | 36 |
| 10/16/2019 | 8 | 16.52 | 40.9 |

Other climate data were collected from North green roof on Javits center.
* Min–max temperature of the day. Data collected from New York City Central Park climate station www.ncdc.noaa.gov.

### 3.2. Chemical indicators

Rain and runoff contained low levels of anions and cations including $Na^+$, $NH_4^+$-N, $K^+$, $Mg^{2+}$, $Ca^{2+}$, $Cl^-$, $NO_2^-$-N, $NO_3^-$-N, $PO_4^{3-}$, and $SO_4^{2-}$. Among these anions and cations, we found $K^+$, $Mg^{2+}$, $Ca^2$, $PO_4^{3-}$, and $SO_4^{2-}$ were significantly higher ($P < 0.05$) in the runoff than in the rainwater (Supplementary Material Table S2). The runoff contained TC (mean = 12.78 mg/L, n = 10) and NPOC (mean = 7.10, n = 10), while rainwater contained 5.11 mg/L (n = 10) of TC and 4.20 mg/L (n = 10) of NPOC. The concentration of $NO_2^-$-N was close to 0 in most of samples in both rainwater and runoff samples.

Because there are no standards or regulations for runoff water quality, in this research we compared our results with standards proposed by the US Environmental Protection Agency (EPA) for freshwater (U.S. Environmental Protection Agency, 1986; U.S. Environmental Protection Agency, 2009), primary and secondary drinking water standards. Among all the anions and cations measured, only phosphate was higher than EPA recommended freshwater standards (Table 2); the others were all below standards or no suggested values.

### 3.3. Microbial indicators

A high abundance of total coliform and *E. coli* were detected in the green roof runoff (Fig. 2, Table 3) significantly higher than found in rainwater. *E. coli* in the runoff exceeded the EPA recreational limit (200 CFU/100 mL) and FDA guidelines (126 CFU/100 mL of geometric mean or 235 CFU/100 mL max). Moreover, the total coliform and *E. coli* concentrations were compared with reclaimed or recycled water allowable agricultural use standards adopted in different states, including Arizona, California, Colorado, Delaware, Florida, Hawaii, Idaho, Minnesota, New Jersey, North Carolina, Virginia and Washington State (Rock et al., 2019). In general, a higher limit is set for non-food crops than for food crops which come into direct contact with humans. In addition, non-food crops irrigation use requirements were found to be stricter than the federal EPA recreational limit in most states. Although our study measured total coliform and not fecal coliform counts (but assuming they are correlated), the runoff from the green roof

**Table 2**
Water quality chemical indicator. For each event, at least one rain sample was collected from each rain sampling device and 4–12 drain samples were collected. TOC and HPLC measured each sample triplicates.

| | EPA freshwater criteria, primary or secondary drinking water standards mg/L | Sample type | Number of event | Mean | Exceed limit |
|---|---|---|---|---|---|
| pH | 6.5–8.5 | Rain | 3 | 6.96 | No |
| | | Runoff | 4 | 7.39 | No |
| Conductivity | – | Rain | 8 | 38.95 | No |
| | | Runoff | 8 | 290.51 | No |
| $Na^+$ | – | Rain | 10 | 1.18 | No |
| | | Runoff | 10 | 2.00 | No |
| $NH^+$-N | – | Rain | 10 | 0.47 | No |
| | | Runoff | 10 | 0.21 | No |
| $K^+$ | – | Rain | 10 | 0.25 | No |
| | | Runoff | 10 | 1.44 | No |
| $Mg^{2+}$ | – | Rain | 10 | 0.13 | No |
| | | Runoff | 10 | 1.40 | No |
| $Ca^{2+}$ | – | Rain | 10 | 0.41 | No |
| | | Runoff | 10 | 12.83 | No |
| $Cl^-$ | 250 | Rain | 10 | 2.76 | No |
| | | Runoff | 10 | 3.70 | No |
| $NO_2^+$-N | 1 | Rain | 10 | 0.19 | No |
| | | Runoff | 10 | 0.03 | No |
| $NO_3^+$-N | 10 | Rain | 10 | 0.30 | No |
| | | Runoff | 10 | 1.25 | No |
| $PO_4^{3-}$ | 0.05 | Rain | 10 | 0.45 | Yes |
| | | Runoff | 10 | 0.57 | Yes |
| $SO_4^{2-}$ | 250 | Rain | 10 | 1.36 | No |
| | | Runoff | 10 | 7.85 | No |
| NPOC | | Rain | 7 | 4.20 | No |
| | | Runoff | 10 | 7.10 | No |
| TC | | Rain | 7 | 5.11 | No |
| | | Runoff | 10 | 12.78 | No |

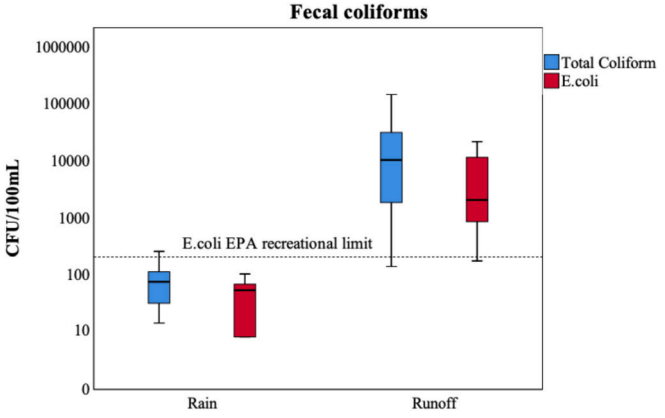


**Fig. 2.** Total coliforms and *E. coli* in rain and runoff samples quantified by Selective media culturing methods.

during the summer (May to October) were still significantly higher than Arizona's and Florida's limit for non-food irrigation (800–4000 CFU/100 mL maximum of fecal coliform), the highest limits adopted by individual states. Although *E. coli* and total coliform counts were high, *Enterococcus* were relatively low in rainwater and runoff samples and were below the EPA recreational limit (33 CFU/100 mL) and reclaimed water treatment and allowable agricultural uses in Virginia (Level 1 Reclaimed water 11 CFU/100 mL geo mean, 24 CFU/100 mL maximum; Level 2 Reclaimed water 35 CFU/100 mL geo mean, 104 CFU/100 mL maximum) (LIS, 2018).

Results from qPCR analyses targeting *Campylobacter* spp., *Salmonella* spp., *Legionella* spp., *Mycobacterium* spp., and *Catellicoccus marimammalium* are presented in Fig. 3. These analyses showed that *Mycobacterium* was relatively low but always detected in runoff samples (n = 9) and rainwater samples (n = 9). *Campylobacter* (a foodborne pathogen) was relatively low or below detection in both rain (n = 9) and runoff samples (n = 9). Similarly, *Salmonella* was relatively low or below detection in rain samples (n = 9) but always detected in runoff samples (n = 9). In contrast, *Legionella* was found in higher concentrations in runoff samples (n = 9) than rain samples (n = 9). Across all sampling events, runoff samples contained higher amount of *Mycobacterium* spp. and *Legionella* spp. than the rainwater samples (Independent *T*-test, *Mycobacterium* $P = 0.036$; *Campylobacter* $P = 0.494$; *Salmonella* $P = 0.213$; *Legionella* $P = 0.026$) (Fig. 4).

The qPCR results for the gull-associated markers, targeting *C. marimammalium* indicated its presence in all runoff samples and rain samples in two of the rain events from May 2019 to October 2019, but too low to be detected in the three events in 2018.

## 4. Discussion

Although recycled runoff from green roofs would not likely be used be as potable drinking water, previous studies have compared the water quality of runoff from green roofs with United States Environmental Protection Agency (EPA) primary and secondary drinking water standards (Mendez et al., 2011) and EPA recommended freshwater standards (Vijayaraghavan et al., 2012). In New York City, recycling of treated wastewater and harvested rainwater must comply with the water quality standards of NYC plumbing code; acceptable uses are toilet and urinal flushing, cooling tower makeup, washing of sidewalks, streets or buildings, laundry, subsurface or drip landscape irrigation systems, hose irrigation, or other approved uses that are located in the same lot as the water recycling system itself (NYC Department of Buildings, 2014). Moreover, other water standards such as recreational water limit (U.S. Environmental Protection Agency, 2012), reclaimed or recycled water for agricultural can be applied if reuse of the runoff water is not for drinking purposes but that may cause individuals to come in contact with the water (e.g., occupationally through maintenance of cooling water or via irrigation activities). To date, there are no Federal microbial indicator standards set for irrigation waters, rather water reuse regulations in the United States are developed at the state and local levels (Rock et al., 2019).

The level of chemical components analyzed in the rainwater and runoff collected from the Javits green roof in New York City from 10 rain events from Sept 2018 to Oct 2019 was relatively low. Even though the

**Table 3**
Total coliforms and *E. coli* in rainwater and runoff water, and the number of herring gulls present on the roof were counted between 1 and 10 days of the sample collection date.

| Sampling date | | 9/25/18 | 10/11/18 | 11/5/18 | 5/13/19 | 5/28/19 | 6/18/19 | 7/17/19 | 7/23/19 | 8/9/19 | 10/16/19 |
|---|---|---|---|---|---|---|---|---|---|---|---|
| *E. coli* | Rain | Not collected | >10 | Not collected | >40 | Not collected | >60 | <10 | >100 | >60 | <10 |
| CFU/100 mL | Runoff | Not collected | >200 | Not collected | >150 | >1000 | >2000 | >10,000 | >20,000 | >12,000 | >600 |
| Total coliform | Rain | Not collected | <10 | Not collected | >100 | Not collected | >50 | >10 | >250 | >30 | >100 |
| CFU/100 mL | Runoff | Not collected | >500 | Not collected | >100 | >1000 | >2000 | >10,000 | >39,000 | >140,000 | >23,000 |
| Herring gulls present | | 29 | 3 | Not collected | 98 | 186 | 219 | 148 | 204 | 132 | 1 |

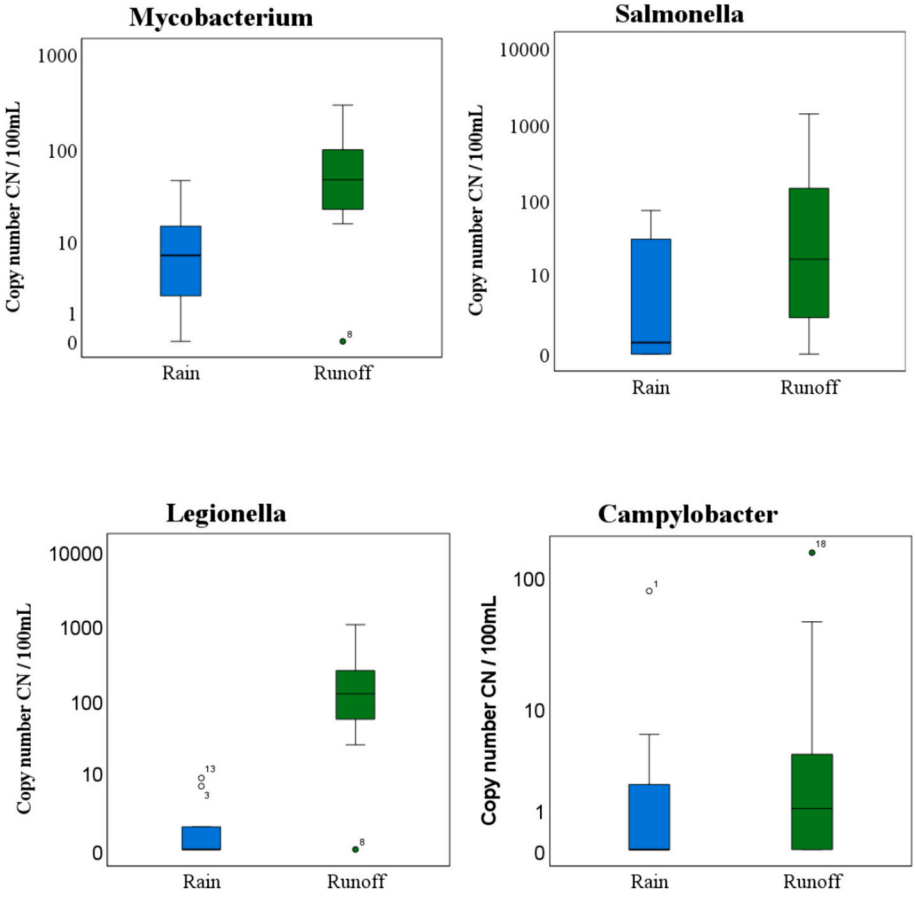


**Fig. 3.** *Mycobacterium*, *Salmonella*, *Legionella* and *Campylobacter* quantified by qPCR in both rainwater and drain water.

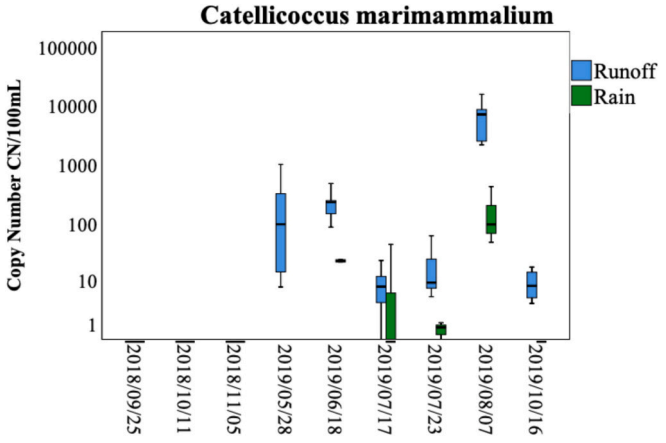


**Fig. 4.** *Catellicoccus marimammalium* concentration in rainwater and runoff water in each rain event. Rain sample in 2019/5/28 was not collected.

runoff contained significantly higher concentrations of anions and cations than rainwater, the level of analytes measured in the runoff water were below EPA primary and secondary drinking water standards, except for phosphorus (Table 2). The green roof runoff had more TC and NPOC than rainwater, meaning the green roof acts as a source of leachable dissolved organic compounds and carbonates. The high concentration of phosphorus could be due to leaching of fertilizers previously applied to the green roof (Berndtsson et al., 2006; Teemusk and Mander, 2007). Ammonium nitrogen was found in both the rain and runoff in this study. Ammonia is very soluble in water and could also have originated in prior fertilization of the green roof. It could also be from fecal droppings of birds or from atmospheric deposition. In rural settings, the main source of ammonia in air is from agriculture includes fertilization and livestock (Behera et al., 2013), while automobiles and industry are also dominant urban sources of ammonia (Behera et al., 2013; Liu et al., 2014). Green roofs have previously been shown to be a sink for ammonia (Gregoire and Clausen, 2011), and our work supports this notion since the runoff did not contain more ammonia than rainwater ($P = 0.224$). High nutrient levels found in urban stormwater are commonly a concern when considering their recycling or reuse potential (Czemiel Berndtsson, 2010). Prior studies on green roof water quality have shown that inorganic pollutants, total suspended solids (TSS), and trace metals (Banting et al., 2005; Gregoire and Clausen, 2011; Morgan et al., 2013) can be reduced by green roofs, but that the use of fertilizers and leaching from the growing media could introduce nitrogen, organic carbon, and phosphorus into the runoff (Berndtsson et al., 2006; Buffam and Mitchell, 2015; Burszta-Adamiak, 2020; Pęczkowski et al., 2020; Zhang et al., 2020). In addition, airborne pollutants can adhere to plant surfaces, which can become entrained in falling precipitation, leading to their presence in water that infiltrates the growing media or that runs off the green roof into roof drains (Liu et al., 2019; Rodak et al., 2020; Taguchi et al., 2020). Beyond leaching organic carbon and nutrients (N and P), other researchers have found that green roofs can increase the pH of mild acid rain from pH 5–6 to over 7–8 (Berndtsson et al., 2006; Teemusk and Mander, 2007; Bliss et al., 2009). In this study, we did not find the rain to be more acidic than the runoff, but we found that the rain had a higher pH range than the runoff. Constituents within the green roof, like carbonates and phosphates, that become dissolved in the runoff can act as a buffer stabling pH variation (Berghage et al., 2007).

The most significant finding focused on the microbiological components of the runoff from the Javits Center green roof. Previous studies have shown that *E. coli* can be found in runoff from green roofs (Van

Seters et al., 2009). *E. coli* is a facultative anerobic bacteria commonly found in the intestines of warm-blooded animals. This trait, along with its ease of cultivation has led to its common use as a Fecal Indicator Bacteria (FIB), where its presence often indicates the presence of fecal contamination as well as other waste-associated pathogens (Ahmed et al., 2019). Since *E. coli* generally does not survive well outside of the intestinal tract, high amounts of *E. coli* in runoff means recent fecal contamination occurred on the green roof or drainage pipe.

In 2019 up to 219 herring gulls were observed on the green roof, the majority of which were actively nesting and introducing a source of potential contamination. Herring gull activity peaked in June and July of 2019, coinciding with the observation of high concentration *E. coli* in the runoff water. The presence of *C. marimammalium*, a gull-associated marker, in runoff samples in all events from May 2019 to October 2019 indicates that gull droppings were the likely source of pathogens. *E. coli* and total coliforms were also more abundant in the events with detectable *C. marimammalium*. These findings suggest that fecal contamination detected in the runoff from the green roof likely results from the large number of gulls on the green roof. However, the location where the runoff water was collected was half green and half conventional, and while contamination from an active gull colony was evident, future research should examine how roof with conventional and green roof components affect bacterial contamination of the roof runoff. For example, we suspect that due to herring gull behavior on the Javits green roof, the location sampled may have had some of the highest concentration of feces on the roof. The elevated part of the roof where we sampled is an area where, based on observations, an abundance of herring gulls would congregate and loaf, likely due to the high vantage point. Thus, the fecal contamination detected may be higher than other areas on the green roof. Further studies are needed to determine the variability of fecal contamination at a finer scale at different green and conventional sections of the roof. In addition, the peak in fecal bacteria indicators appeared after the peak of gull numbers (Table 3). This suggests potential factors requiring further investigation. Possible hypotheses include reduced rainfall in the fall leading to less roof flushing, shifts in gull behavior such as increased loafing time on the roof, or the onset of dormancy in *Sedum* plants, potentially altering roof characteristics.

Despite the high levels of fecal indicators found in the runoff water from the green roof, the levels of potential opportunistic pathogens, including *Campylobacter*, *Salmonella*, and *Mycobacterium* were low except for *Legionella* spp. The qPCR primers used for *Legionella* spp. were designed to detect all species in this genus, not just the *Legionella pneumophila*, which can cause Legionnaire's disease in humans. Since *Legionella* are commonly found in moist environments, as well as water distribution systems of large buildings (Lin et al., 1998), it is not surprising that they are highly abundant in the runoff from the green roof. Further studies that specifically target *L. pneumophila*, either via culturing or qPCR methods, are needed to determine if the high levels of *Legionella* spp. detected in this study represent a potential human risk.

## 5. Conclusions

Overall, in this study we found large amounts of fecal contamination in the runoff water from the Javits Center green roof. *E. coli* in runoff water collected on Javits Center roof ranged from 150 to over 20,000 CFU/100 mL and total coliform values ranged from 100 to over 140,000 CFU/100 mL, which were in exceedance of the NYC plumbing code for recycled water (2.2 CFU/100 mL *E. coli* and 100 CFU/100 mL total coliform), the EPA recreational limit (200 CFU/100 mL *E. coli*), the FDA guidelines (126 CFU/100 mL of geometric mean or 235 CFU/100 mL maximum *E. coli*) and the reclaimed or recycled water allowable agricultural use standards in some states (800–4000 CFU/100 mL maximum fecal coliform). On the other hand, chemically the water quality of the runoff water (specifically of $Cl^-$, $NO_2^-$-N, $NO_3^-$-N and $SO_4^{2-}$) were below US EPA drinking water standards. Despite the high concentrations of total coliform measured in the runoff, the levels of potential opportunistic pathogens like *Campylobacter*, *Salmonella*, and *Mycobacterium* were low, except for *Legionella* spp. However, since the primers used to quantify *Legionella* spp. did not specifically target *L. pneumophila*, which is the primary agent for Legionnaires Disease, further studies are needed that quantify *L. pneumophila* via culturing methods, such as plate culture, qPCR, Most Probable Number Methods (Walker and McDermott, 2021), to determine if levels in the green roof runoff represent a potential human risk.

This study found that the fecal contamination on the Javits Center green roof was likely due to the large number of birds, especially gulls, which have come to nest and live on the green roof of Javits from May to October. During this period, in addition to high concentrations of *E. coli* and total coliform, *C. marimammalium*, a gull-associated marker, was detected frequently via qPCR in runoff, indicating that gull droppings are the likely contributing source to the large amount of fecal contamination found in the runoff water from the Javits green roofs during the summers studied.

Any runoff generated from the Javits Center roof is discharged to the local combined sewer system. However, this study suggests that if the runoff from this green roof, or another one populate by gulls, were to be recycled for irrigation, it is likely that treatment would be required. This treatment would aim to lower bacterial levels, potentially to meet the EPA's recreational limit for *E. coli* (200 CFU/100 mL) and the maximum fecal coliform standards for reclaimed or recycled water, which vary across states, ranging from 800 to 4000 CFU/100 mL. In relation to the high concentration of *E. coli* and total coliform observed in this study, in order to reduce the fecal contamination, disinfection may be needed (Marsalek and Rochfort, 2004), in combination with methods to reduce the gull populations. For example, previous research showed that specially trained dogs could reduce gulls on recreational beach by a 50 %, resulting in a 38 % and 29 % decrease in *Enterococcus* spp. and *E. coli* densities, respectively (Nevers et al., 2018). However, gull control activities do not always reduce the number of gulls in an area. Rather, gulls often just move to a nearby nesting location. In urban areas disturbed gulls would likely move to a nearby roof, and likely a conventional roof, considering how few green roofs are in most cities. Further research is needed to determine if runoff from green roof gull colonies differs chemically and biologically from runoff from gull colonies on conventional roofs, but in either case, if gull colonies are present on a roof, water quality should be assessed if the runoff is being considered for use. Gulls are nesting on both green and conventional roofs in New York City. Of the three known major gull colonies in NYC, two are on conventional roofs (Rikers Island, Governors Island) and one is on a green roof (Javits). This work establishes an important baseline. Gull colonies in NYC are unavoidable, but green roofs likely lessen their water quality impact. Future work comparing gull-induced pollution in green roof runoff with gull-induced pollution in conventional roof runoff will be very telling. For instance, if green roofs exhibit lower fecal coliform levels than conventional roofs with similar gull populations, it may suggest that green roofs can attenuate bacterial pollution originating from gulls.

## CRediT authorship contribution statement

**Jinjie He:** Formal analysis, Investigation, Writing – original draft, Writing – review & editing. **Elrod Owusu-Asumeng:** Investigation. **Kate Zidar:** Investigation. **Julian Stolper:** Investigation. **Sudipti Attri:** Writing – original draft. **Jacob R. Price:** Investigation, Methodology. **Dustin Partridge:** Investigation, Writing – original draft. **Franco Montalto:** Project administration, Supervision. **Christopher M. Sales:** Conceptualization, Supervision.

## Declaration of competing interest

Franco Montalto reports financial support was provided by Jacob K. Javits Center.

## Data availability

Data will be made available on request.

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## Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.scitotenv.2024.174430.

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