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DATA PAPER

Seasonal Abundance of Fecal Indicators and Opportunistic Pathogens in Roof-Harvested Rainwater Tanks

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Here we provide seasonal data on the concentrations of total coliform, *Escherichia coli* and *Enterococcus* spp. and six opportunistic pathogens (*Acanthamoeba* spp., *Legionella* spp., *Legionella pneumophila*, *Mycobacterium avium*, *Mycobacterium intracellulare*, and *Pseudomonas aeruginosa*) of public health significance in 24 tank water samples over six monthly sampling events from August 2015 to March 2006. Quantitative PCR (qPCR) assays were chosen for the quantification of six opportunistic pathogens and culture-based methods were used for the enumeration of fecal indicators. The data fle has been stored in a publicly available repository. The data on concentrations of opportunistic pathogens in RHRW will provide information for rainwater users regarding potential seasonality of risks. Quantitative data presented in this study can be used to perform quantitative microbial risk assessment (QMRA) of RHRW for various potable and nonpotable uses. Data can be used by health regulators to develop guidelines related to RHRW.

Keywords: Roof-harvested rainwater; opportunistic pathogens; faecal indicator bacteria

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1. Overview

Introduction/Study Description

Roof-harvested rainwater (RHRW) is currently being used globally to supplement potable and nonpotable water supplies. Pathogens can enter into the tanks through aerosol deposition, plant litter, and animal fecal matter via roof runoff. Case-control studies have reported some associations between untreated rainwater consumption and gastroenteritis based on enteric pathogens [1]. Improvements in disinfection practices for centralized drinking water systems have reduced the disease burden of enteric pathogens in some regions. Because of this, increased attention has been devoted to opportunistic pathogens that grow in biofilms on the surfaces of distribution system and premise plumbing pipes [2]. Opportunistic pathogens cause illness primarily for immunocompromised people [3]. A high abundance of opportunistic pathogens in tank water samples has been reported [4, 5, 6] supporting the need to consider potential health risks. However, most studies in the research literature reported the occurrence/abundance of opportunistic pathogens based on testing a sample at a single time-point from a tank. Therefore, there is a paucity of information available regarding seasonal or temporal variations in opportunistic pathogen occurrence and abundance. Here we provide seasonal data on the concentrations of fecal indicators (total coliform, *Escherichia coli* and *Enterococcus* spp.) and six opportunistic pathogens (*Acanthamoeba* spp., *Legionella* spp., *Legionella pneumophila*, *Mycobacterium avium*, *Mycobacterium intracellulare*, and *Pseudomonas aeruginosa*) of public health significance in tank water over six monthly sampling events. qPCR methods were chosen for the quantification of six opportunistic pathogens and culture-based methods were used for the enumeration of three fecal indicators.

2. Context

Spatial coverage

Brisbane and Currumbin Ecovillage, Southeast Queensland, Australia

Temporal coverage

18/08/2015–11/03/2016

Species

Total coliforms
Escherichia coli
Enterococcus spp.
Legionella spp.

Legionella pneumophila
Acanthamoeba spp.
Mycobacterium avium
Pseudomonas aeruginosa
Mycobacterium intercellulare

3. Methods

Quantitative qPCR assays were used to measure the concentrations of six opportunistic pathogens in 24 RHRW tanks over six months. In addition, total coliform, *E. coli* and *Enterococcus* spp. were enumerated using culture-based methods.

Sampling strategy

Monthly water samples were collected from 24 roof-harvested rainwater tanks on six separate events from August 2015 to March 2016, giving a total number of 144 tank water samples. The tap/spigot connected directly to the rainwater tank was wiped with 70% ethanol, and the stored water was run for 15 s prior to filling a 10 L sterile container. Samples were immediately transported to the laboratory, kept at 4°C, and processed within 6–12 h.

Steps

Colilert and Enterolert (IDEXX Laboratories, Westbrook, Maine, USA) Test kits were used to determine the concentrations of FIB (total coliforms, *E. coli*, and *Enterococcus* spp. in 100 mL of each tank water sample. For the measurements of opportunistic pathogens in tank water samples, a 10 L water sample from each rainwater tank was concentrated by a hollow-fiber ultrafiltration system using Hemoflow FX 80 dialysis filters (Fresenius Medical Care, Bad Homburg, Germany) as previously described [6]. The concentrated sample was filtered through a 0.45 μm cellulose filter paper (Advantec, Tokyo, Japan), and stored at –80°C until DNA extraction. DNA was extracted using a PowerSoil Max DNA Kit (MO BIO, Carlsbad, California, USA) according to the manufacturer's instructions. qPCR assays were performed using previously published primers, probes, and optimized reaction mixtures and cycling parameters [7].

Standard curves for all qPCR assays were constructed using synthesized plasmid DNA Integrated DNA Technologies, Coralville, IA, U.S.A.). The purified plasmid DNA was serially diluted to create a standard ranging from 1×10^6 to 1 gene copies per μL of DNA. For each standard, the genomic copies were plotted against the cycle number at which the fluorescence signal increased above the quantification cycle value (C_q value). All qPCR reactions were performed in triplicate using a Bio-Rad CFX96 thermal cycler. The qPCR method limit of quantification (MLOQ) was also determined from the C_q values obtained for each standard. The minimum concentration of copies from the standard series detected in 3/3 qPCR replicated was considered qPCR LLOQ. The dilution below this series was considered the method limit of detection (MLOD).

Quality Control

Method and reagent blank runs were included during the sample processing steps to ensure to detect carry over contamination. No carryover contamination was observed. To minimize qPCR contamination, DNA extraction and qPCR setup were performed in separate laboratories. The minimum information for publication of quantitative real-time PCR experiments (MIQE) guidelines were used throughout the qPCR quantification of opportunistic pathogens [8].

Constraints

N/A

Privacy

Each roof-harvested rainwater tank was designated to a unique ID (T1, T2, T3 and T4 etc.) to to anonymise the data.

Ethics

N/A

4. Dataset description

Object name

Seasonal_data_FIB_opportunistic_pathogens_RHRW.xlsx

Data type

Primary and processed data.

Ontologies

N/A

Format names and versions

Excel

Creation dates

18/08/2015–11/03/2016

Dataset creators

Ahmed W, Hamilton KA, Toze S, Haas CN

Language

English

Programming language

N/A

Licence

Creative Commons Attribution 4.0 International Licence

Accessibility criteria

N/A

Repository location

https://doi.org/10.4225/08/58771be56f814

Publication date

12/01/2016

5. Reuse potential

The data on concentrations of opportunistic pathogens in RHRW will provide information for rainwater users regarding potential seasonality of risks. Quantitative data presented in this study can be used to perform quantitative microbial risk assessment (QMRA) of RHRW for various potable and nonpotable uses. Data can be used by health regulators to develop guidelines related to RHRW.

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Competing Interests

The authors have no competing interests to declare.

References

1. **Merritt, A, Miles, R** and **Bates, J.** 1999. An outbreak of *Campylobacter enteritis* on an island resort, north Queensland. *Communicable Diseases Intelligence*, 23(8): 215–219.
2. **Pruden, A, Edwards, MA** and **Falkinham, JO, 3rd.** 2013. State of the Science and Research Needs for Opportunistic Pathogens in Premise Plumbing. Denver, CO: Water Research Foundation.
3. **Falkinham, JO, 3rd, Hilborn, ED, Arduino, MJ, Pruden, A** and **Edwards, MA.** 2015. Epidemiology and ecology of opportunistic premise plumbing pathogens: *Legionella pneumophila*, *Mycobacterium avium*, and *Pseudomonas aeruginosa*. *Environmental Health Perspective*, 123(8): 749–758. DOI: https://doi.org/10.1289/ehp.1408692
4. **Ahmed, W, Brandes, H, Gyawali, P, Sidhu, JPS** and **Toze, S.** 2014. Opportunistic pathogens in roof-captured rainwater samples, determined using quantitative PCR. *Water Research*, 53: 361–369. DOI: https://doi.org/10.1016/j.watres.2013.12.021
5. **Dobrowsky, PH, De Kwaadsteniet, M, Cloete, TE** and **Khan, W.** 2014. Distribution of indigenous bacterial pathogens and potential pathogens associated with roof-harvested rainwater. *Applied and Environmental Microbiology*, 80(7): 2307–2316. DOI: https://doi.org/10.1128/AEM.04130-13
6. **Hamilton, K, Ahmed, W, Palmer, A, Sidhu, JPS, Hodgers, L, Toze, S** and **Haas, CN.** 2016. Public health implications of *Acanthamoeba* and multiple potential opportunistic pathogens in roof-harvested rainwater tanks. *Environmental Research*, 150: 320–327. DOI: https://doi.org/10.1016/j.envres.2016.06.017
7. **Hamilton, KA, Ahmed, W, Palmer, A, Smith, K, Toze, S** and **Haas, CN.** 2017. Seasonal assessment of opportunistic premise plumbing pathogens in roof-harvested rainwater tanks. *Environmental Science & Technology* (accepted). DOI: https://doi.org/10.1021/acs.est.6b04814
8. **Bustin, SA, Benes, V, Garson, JA, Hellemans, J, Huggett, J, Kubista, M, Mueller, R, Nolan, T, Pfaffl, MW, Shipley, GL, Vandesompele, J** and **Wittwer, CT.** 2009. The MIQE guidelines: minimum information for publication of quantitative real-time PCR experiments. *Clinical Chemistry*, 55: 611–622. DOI: https://doi.org/10.1373/clinchem.2008.112797

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