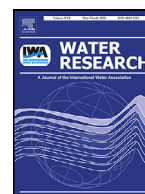




Since January 2020 Elsevier has created a COVID-19 resource centre with free information in English and Mandarin on the novel coronavirus COVID-19. The COVID-19 resource centre is hosted on Elsevier Connect, the company's public news and information website.

Elsevier hereby grants permission to make all its COVID-19-related research that is available on the COVID-19 resource centre - including this research content - immediately available in PubMed Central and other publicly funded repositories, such as the WHO COVID database with rights for unrestricted research re-use and analyses in any form or by any means with acknowledgement of the original source. These permissions are granted for free by Elsevier for as long as the COVID-19 resource centre remains active.



Screening Level Risk Assessment (SLRA) of human health risks from faecal pathogens associated with a Natural Swimming Pond (NSP)

Susan Petterson^{a,b,*}, Qiaozhi Li^c, Nicholas Ashbolt^{c,d}

^a Water & Health Pty Ltd, North Sydney, NSW 2060, Australia

^b School of Medicine, Griffith University, Gold Coast QLD 4222, Australia

^c School of Public Health, University of Alberta, Edmonton, Alberta T6G 1C9, Canada

^d Southern Cross University, Lismore, NSW 2480, Australia



ARTICLE INFO

Article history:

Received 27 May 2020

Revised 1 October 2020

Accepted 5 October 2020

Available online 5 October 2020

Keywords:

QMRA

Recreational water quality

Cryptosporidium, *Norovirus*, *Campylobacter*,

Natural swimming ponds

ABSTRACT

Natural swimming ponds (NSPs) are artificially created bodies of water intended for human recreation, characterised by the substitution of chemical disinfection with natural biological processes for water purification. NSPs are growing in popularity, however little is known regarding the public health risks. A screening level risk assessment was undertaken as an initial step in assessing the first Canadian public NSP located in Edmonton, Alberta. Risk of enteric pathogens originating from pool bathers was assessed under normal conditions and following accidental faecal release events. The performance of the natural treatment train for health protection was quantified with and without the addition of UV disinfection of naturally-treated water, and compared to the US EPA benchmark to provide a reference point to consider acceptability. Estimated levels of pathogen contamination of the pond were dependant upon the discrete number of shedders present, which in turn depended upon the prevalence of infection in the population. Overall performance of the natural disinfection system was dependant upon the filtration rate of the natural treatment system or turnover time. Addition of UV disinfection reduced the uncertainty around the removal efficacy, and mitigated the impact of larger shedding events, however the impact of UV disinfection on the natural treatment biome is unknown. Further information is needed on the performance of natural barriers for pathogen removal, and therefore challenge studies are recommended. Given the identified risks, the pool is posted that there is risk from accidental faecal releases, as in any natural water body with swimmers. Screening level risk assessment was a valuable first step in understanding the processes driving the system and in identifying important data gaps.

© 2020 Elsevier Ltd. All rights reserved.

1. Introduction

Natural swimming ponds (NSPs) are artificially created bodies of water intended for human recreation, characterised by the substitution of chemical disinfection with natural biological processes for water purification. The first NSP in recent times was built in Austria in the early 1980s, and by 2010 more than 20,000 NSPs had been constructed of which a hundred were open to the public (Littlewood 2005) cited in (Casanovas-Massana and Blanch 2013). Despite the growing popularity of these natural systems, little is known regarding the adequacy of the natural processes for enteric pathogen removal to provide adequate water quality for swimmers. Also, it is well known by health authorities that most recreational disease is not identified in normal health surveillance programs

(Fewtrell and Kay 2015), hence the majority go unreported even though there may be significant impacts on society including lost work days (Dwight et al., 2005). In general, the chlorine-resistant parasitic protozoan, *Cryptosporidium hominis* is the leading cause of gastroenteritis in swimming pools, and results from faecal accidents/releases (Suppes et al., 2016) referred to in this paper as bather shedding. Human faeces may contain pathogens and is a known pathway of infection in recreational water environments (Chalmers 2012; Dale et al., 2010; Graciaa et al., 2018; Pond 2005). Hence, in a natural pool, without chemical disinfectant residual, expected bather shedding of pathogens will go untreated until the pool water is passed through sufficient 'natural' barriers, with human enteric viruses the most numerous and infectious of these enteric pathogens (Ashbolt 2015).

While the social and health benefits associated with recreational water environments are well recognized, various outbreaks have been associated with microbiological contamination of recreational waters in natural ponds and lakes (Blostein 1991;

* Corresponding author.

E-mail address: s.petterson@waterandhealth.com.au (S. Petterson).

Paunio et al., 1999; Sinclair et al., 2009) and inadequately disinfected swimming pools (Barna and Kádár 2012; Sinclair et al., 2009). Hence, the efficacy and reliability of disinfection treatments (natural or artificial) is central to maintaining the microbial safety of recreational waters.

The design and operation of the first Canadian NSP in Edmonton was the focus of this microbial risk assessment. The NSP treatment system designed by Polyplan Kreikenbaum Group GMBH (www.polyplan-umwelt.de) was part of a municipal pool upgrade. As part of the planning process, potential faecal pathogen risks were evaluated, using a Quantitative Microbial Risk Assessment (QMRA) framework. It is recommended to undertake QMRA at increasing levels of detail referred to as a tiered approach (WHO 2016), beginning simply and only increasing complexity as needed. This paper documents the first Screening Level Risk Assessment of pathogen risk undertaken of a NSP. The objective of the screening level assessment, was to characterize the system in a very simplistic way, based on average flow rates (within a simple box flow model) and using available literature data assess factors driving illness risks from accidental ingestion of enteric viruses, bacteria and parasitic protozoa, and to identify future data collection needs for the purpose of quantify risks adequately to support health protection.

2. Materials and methods

The NSP is part of a redevelopment of a public pool at Borden Park, Edmonton, Canada (https://www.edmonton.ca/activities_parks_recreation/borden-park-outdoor-pool.aspx). The design consists of a large rectangular main pool, a shallow children's pool (referred to as kiddie pool), and an area of floor nozzles designed for children's water play. The water flow is connected between all pools (see Fig. 1) via the pump well (B1) and is directed to external filtration units (Neptune filter, Hydrobotanic and submerse filters) for purification. The public pool is fenced from wildlife, and fed by potable water from the mains drinking water supply, therefore the significant only source of faecal contamination considered in the assessment was from bathers.

The first step in any QMRA is to undertake a problem formulation, defining clearly the purpose and scope of the assessment (WHO 2016). To address each class of microbial pathogen (viral, bacterial, parasitic protozoan) for the first tier in the QMRA process, the following reference pathogens were selected to represent

each microbial group: *Norovirus*, *Campylobacter jejuni* and *Cryptosporidium hominis*; with *Norovirus* and *C. jejuni* representing some of the most prevalent pathogenic enteric viruses and bacteria reported in Edmonton sewage (Banting et al., 2016; Qiu et al., 2015) and known to infect recreational swimmers (Guy et al., 2018); *C. hominis* was selected as oocysts are known to be far more resistant to environmental decay processes than *Giardia* cysts (Hamilton et al., 2018). Risk to adults and children were considered separately and compared with the gastrointestinal benchmark of the U.S. EPA for freshwaters of 35 cases of gastroenteritis per 1000 swimming events (EPA, 2012). Exposure scenarios included in the risk assessment were for unintentional shedding during normal operation (nominal load = 252 adults and 17 children; high bather load estimated as 1.5 times the nominal value = 378 adults and 26 children) and larger faecal release events.

First the magnitude of pathogen contamination due to unintentional bather shedding was estimated; and the level of treatment required in order to achieve the benchmark risk was quantified. The capacity of the designed filtration system for treating the pathogen load at the benchmark risk level was then assessed, including the system response to larger pathogen release incidents. The overall objective of the assessment was to assess the adequacy of the proposed treatment system for managing pathogen risks to bathers, and to identify future research needs to better characterize safety.

2.1. Magnitude of pathogen contamination due to unintentional bather shedding

Enteric pathogens are transmitted by the faecal-oral route, and therefore the presence of pathogens in the water column is caused by shedding of faecal material by swimmers. In this model, the pathogen loading under typical operating conditions was estimated as $N \cdot f_s \cdot c_{rp}$ where N is the discrete number of people visiting the facility per day infected with a reference pathogen, f_s is the amount of faecal material (grams) shed per person per day, and c_{rp} is the concentration of reference pathogens in the faeces of infected individuals ($\text{organisms} \cdot \text{g}^{-1}$).

Number of people shedding: Only infected people excrete pathogens. The point prevalence of each of the reference pathogens was estimated in order to model the expected number of excretors present at the pool by day. The point prevalence was estimated from the reported number of cases by week for the

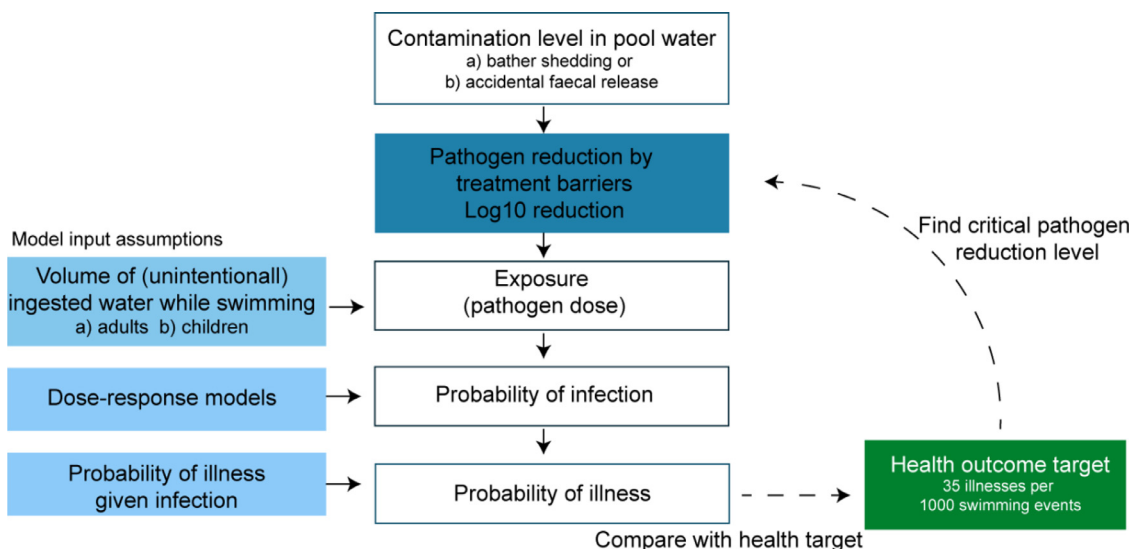


Fig. 1. QMRA approach for quantifying the required pathogen reduction to achieve safety.

Table 1
Input values for estimating the point prevalence for reference pathogen shedding.

	Campylobacter	Norovirus	Cryptosporidium
Reported cases by week*	40	20	15
Under reporting factor	27.2 (Thomas et al., 2013)	288 (Tam et al., 2012)	48.5 (Thomas et al., 2013)
Mean duration of excretion (days)	21 (Havelaar et al., 2009)	28.5 (Tu et al., 2008)	30 (Stehr-Green et al., 1987)
Asymptomatic infection rate	0.8 (Black et al., 1988)	0.3 (Teunis et al., 2008)	0.3 (USEPA 2006)
Calculated Point Prevalence (%)	0.39	0.80	0.11

* A conservative value for the summer season from Alberta Health Services (Dr Lilly Pang pers comm).

province of Alberta, Canada for each pathogen using the formula: $Point\ prevalence = \frac{R \cdot U \cdot D}{7 \cdot N \cdot (1 - A)}$ where R is the reported number of cases per week, U is the underreporting factor, D is the duration of excretion (days), N is the population of Alberta (4200,000) and A is the asymptomatic infection rate. A summary of inputs is included in Table 1. While *Campylobacteriosis* is a reportable illness in Alberta, *Cryptosporidiosis* and *norovirus* infections are not, and therefore data regarding the identified number of cases is not available for analysis. The selection of a conservative (on the high end of the realistic range) weekly rate of reporting was made via discussions with Alberta Health Services.

The number of visitors shedding (shedders) reference pathogens on any given day will be a discrete number. Given the number of visitors (n), and the point prevalence (pp) of shedding in the general population the discrete number of shedders on any given day was modelled using a binomial distribution with parameters n and pp . A sample of the number of shedders was generated using Monte Carlo simulation (10,000 iterations).

Faecal shedding: Unintentional shedding of faecal material by bathers is widely documented (Elmir et al., 2009; Elmir et al., 2007; Gerba 2000). The amount of faeces shed per person is highly uncertain and was estimated relying on measurements from the literature of faecal indicator concentrations in bathing waters. Details of this analysis is included in supplementary material. Given the uncertainty in shedding mass, a reference distribution to describe the variability in faecal excretion was implemented in the model. A reference distribution is recommended to explicitly represent a plausible range, when there is a lack of data to fully describe a distribution (WHO 2016). We chose a triangular distribution (a distribution often used as a rough modelling tool where the range and most likely value within that range can be estimated (Vose, 2008)) for faecal excretion with a mode equal to 0.6 and lower and upper bounds of 0.06 and 6 gs.

Pathogen shedding density: Reported concentrations of pathogens in the faeces (shedding density) is variable. The shedding density appears to vary between individuals, and over the course of an infection. Bambic et al. (2011) reviewed the published data and reported the following relevant to our chosen reference pathogens: a median *Campylobacter* concentration of 10^7 CFU.g⁻¹, ranging 10^1 – 10^8 ; for *Norovirus* the median was 10^8 copies.g⁻¹, and a range of 10^4 – 10^{10} ; and for *Cryptosporidium* the median was 10^5

oocysts.g⁻¹, ranging 10^3 – 10^6 . The range limits and medians were used to define triangular distributions for the shedding density of each reference pathogen.

Using a model constructed in Mathematica® (Wolfram International, version 11.1) a Monte Carlo simulation (10,000 random samples) was undertaken to obtain a random sample of the concentration of each reference pathogen in the main pool and the kiddie pool, assuming complete mixing. The sensitivity of the estimated concentration to each of the model input variables (number of shedders, pathogen density in faeces and magnitude of faecal shedding by bathers) was evaluating using the Spearman Rank Correlation Coefficient to compare each of the input random samples with the generated concentration sample.

2.2. Assessment of treatment requirements (QMRA)

Quantitative microbial risk assessment (QMRA) was used to assess the amount of treatment required in order to achieve the U.S. EPA benchmark risk. Using the approach illustrated in Fig. 2 and the sample of pathogen concentrations from unintentional bather shedding, the required Log₁₀ reduction was estimated. A random sample representing the variability in the treatment requirements was generated using the pathogen concentration samples described in the previous section. A summary of the model input assumptions is included in Table 2.

2.3. Pathogen reduction capacity of designed natural treatment system

A simple box model was applied, assuming complete mixing within each component of the model over each time step, to evaluate the overall pathogen reduction capacity of the treatment system. Given the flow paths represented in Fig. 1 and the standard and maximum flow rates included in Table 3, together with the assumed Log₁₀ removal capacity of each of the barriers, the change in pathogen concentration over time was estimated.

Five removal barriers were considered in the scoping of the QMRA: a wetland system consisting of zooplankton filtering, a hydro-botanic filter and submerge sand/root filter; a commercial designed Neptune™ surface spray gravel media filter; and UV disinfection (Fig. 1). However, there is very limited published data on

Table 2
Summary of QMRA inputs for evaluating required Log₁₀ reduction.

	Adults		Children
Volume of water (unintentionally) ingested	10 mL (Dufour et al., 2017)	38 mL (Dufour et al., 2017)	
	<i>Campylobacter</i>	<i>Norovirus</i>	<i>Cryptosporidium</i>
Dose-response model: Exact Beta-Poisson Parameters	$\alpha=0.024$; $\beta=0.011$ (Teunis et al., 2005)	$\alpha=0.0044$; $\beta=0.0022$ (Messner et al., 2014)	$\alpha=0.115$; $\beta=0.176$ (Teunis et al., 2002)
Probability of illness given infection P_{ill}	0.2 (Black et al., 1988)	0.7 (Teunis et al., 2008)	0.7 (USEPA 2006)
Risk Benchmark	35 illnesses per 1000 swimming events		
Critical dose (mean number of pathogens)	29.4 (7.7)	7.8 (2.1)	13.4 (3.5)
Adults (children)			

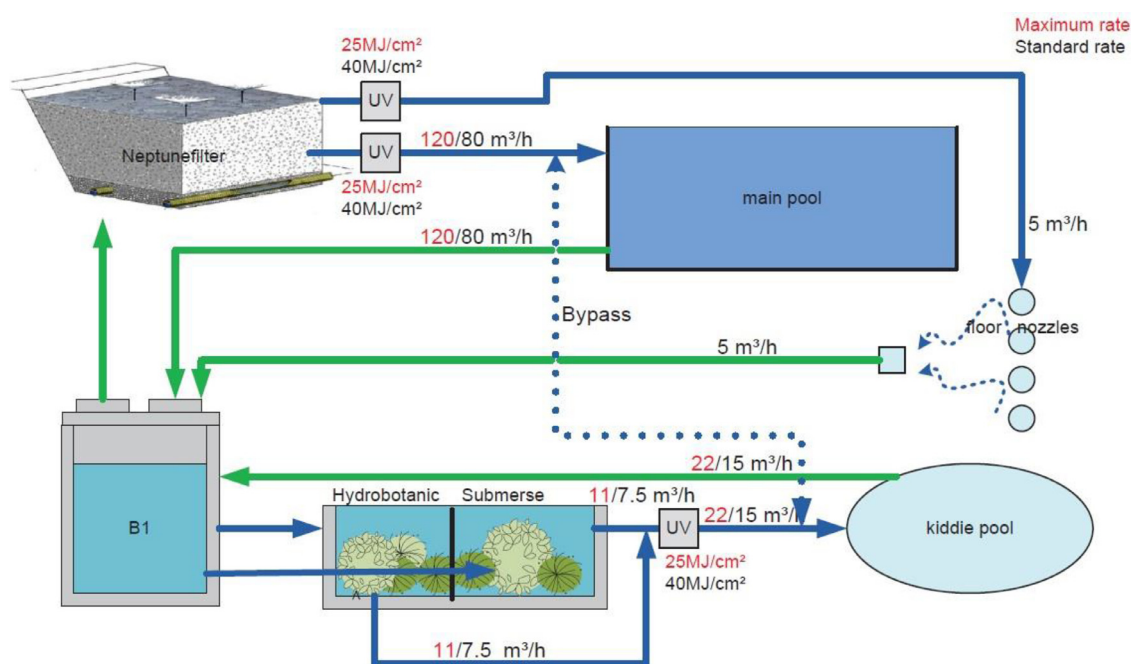


Fig. 2. Simplified water flow chart to provide the basis for the Screening Level QMRA model.

Table 3

Volume, visitor numbers and flow rates to be incorporated into the QMRA model.

		Nominal number of visitors der day*
Volume of Pool (Main Pool) (m ³)	Approx. 885 m ³	252
Volume of Pool (Kiddie Pool) (m ³)	Approx. 60 m ³	17
	Standard rate (m ³ .hour ⁻¹)	Maximum rate (m ³ .hour ⁻¹)
Flow from main pool to B1	80	120
Flow from kiddie pool to B1	15	22
Flow from floor nozzles to B1	5	NA
Flow from B1 to main pool via Neptune Filter	80	120
Flow from B1 to floor nozzles via Neptune Filter	5	NA
Flow from B1 to kiddie pool via hydro botanic plant	7.5	11
Flow from B1 to kiddie pool via submerse substrate filter	7.5	11

* Based on 3.5 m³ per swimmer per visitor per day (Landscaping and Landscape Development Research Society 2011).

Table 4

Best estimate of Log₁₀ removal for treatment barriers within the NSP system (parameters describing a triangular distribution mode [min, max]).

	Best estimate of elimination capacity (log ₁₀ reduction) (with plausible ranges applied in Monte Carlo simulation)		
	Bacteria	Viruses	Protozoa
Zooplankton filtering	0	0	0
Neptune Filter	2 (1, 3)	1 (0.5, 2.5)	1.5 (0.2, 3)
Submerse substrate Filter	1 (0, 2)	0.5 (0, 2)	1 (0.2, 2.5)
Hydro-botanic plant	1 (0, 2)	0.5 (0, 2)	1 (0.2, 2.5)
UV (25 MJ.cm ⁻²)	5	2.6	3

the performance of these NSP barriers (Bruns and Pepper, 2019), so in combination with related literature estimates we provided reasonable point estimate and plausible ranges in Log₁₀ removals for each barrier to be applied within the Screening level QMRA model (Table 4). As such, triangular distributions (defined by mode [min and max]) were selected to describe the variability and uncertainty associated with the Log₁₀ removals.

Zooplankton filtering: Zooplankton grazing is proposed to provide important in-situ disinfection in NSPs (Bruns and Pepper, 2019), yet requires careful consideration as to how well it may be expected to perform as a barrier for protection of human health. Studies have shown that free-living environmental protozoa can

ingest human enteric bacteria and protozoan pathogens including *Cryptosporidium* oocysts (Agasild and Nøges 2005; Connelly et al., 2007; Stott et al., 2003; Trout et al., 2002). Yet very little is known regarding the rate that zooplankton can clear (oo)cysts from the surrounding water, however rates of 22–24 mL-grazer⁻¹.day⁻¹ and 15–19 mL-grazer⁻¹.day⁻¹ for *Cryptosporidium* oocysts and *Giardia* cysts respectively have been reported (Connelly et al., 2007). The filtration capacity of zooplankton reported by (Eydelier and Spieker 2010) cited in (Bruns and Pepper 2019) ranged from 8.5 to 64.8 mL-grazer⁻¹.day⁻¹ for Rotatoria, Copepoda and Cladocera protozoa. The more pressing issue relates to the poorly documented fate of ingested pathogens within zooplankton and their faecal pel-

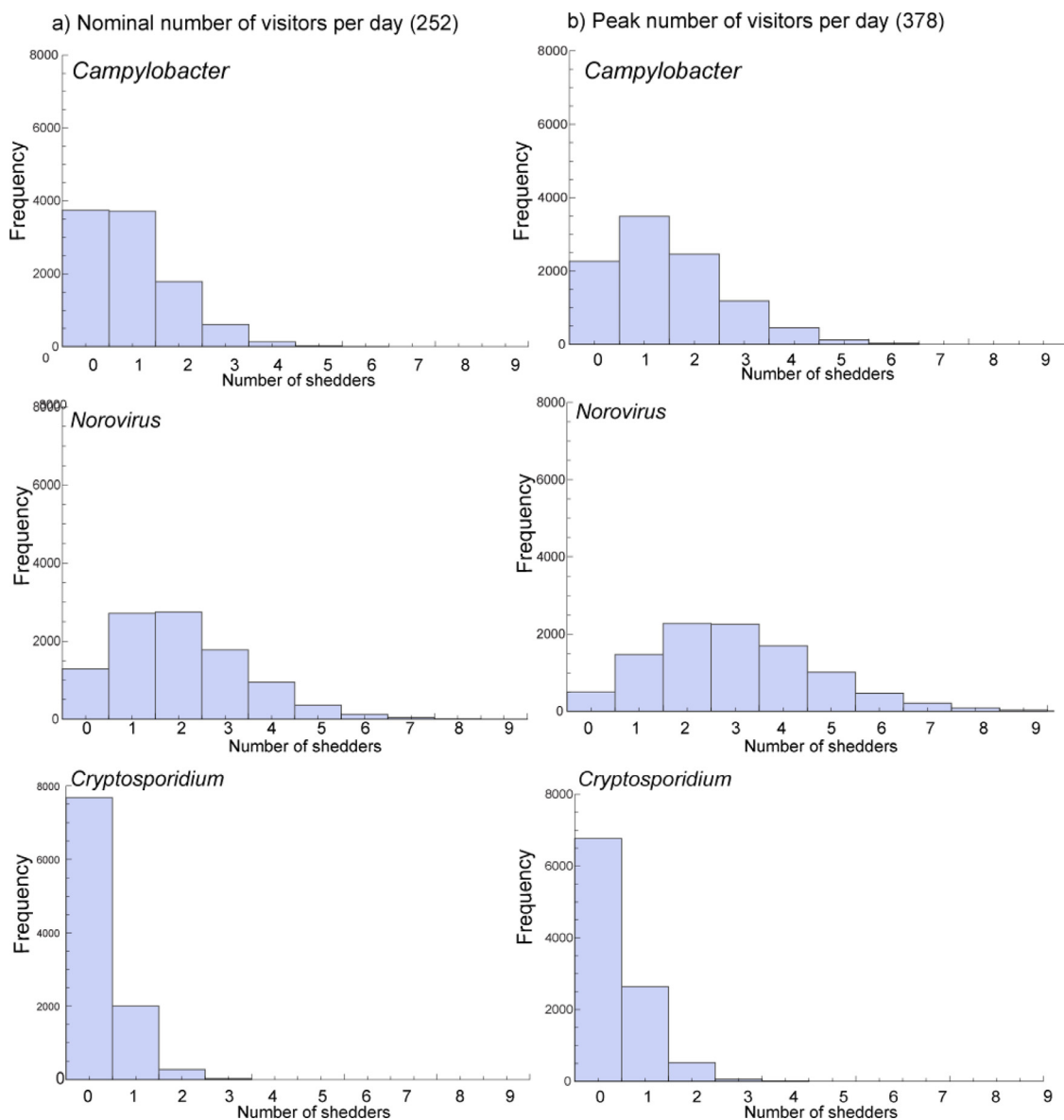


Fig. 3. Frequency histogram of the number of shedders from Monte Carlo sample (10,000 iterations) for the main pool given a) nominal number of visitors and b) peak number of visitors.

lets. Rather than inactivate and digest all ingested pathogens, there is growing evidence that pathogens may be concentrated within zooplankton, where they may be protected from external environmental stressors ultimately favoring their survival (Bichai et al., 2010; Bichai et al., 2014; Bichai et al., 2008; Folkins et al., 2020; Tang et al., 2011). So, while faecal pellets may accumulate within sediments and on plant surfaces, and unknown fraction may stay suspended or be resuspended. For these reasons, zooplankton filtering was not included as a barrier for pathogen removal within this screening-level assessment of the NSP system.

Hydro-botanic and Submerge filter: Two parallel filters were proposed in the design of the NSP including: 1. Hydro-botanic filter with a bed thickness of 2.00 m and a design retention time of 6–8 h; and 2. Submerge filter with a bed thickness of 2.00 m; and a design retention time of 4–5 h.

While these two filters operate in parallel, regarding their likely removal efficiency they are considered here together due to the similarity of mechanisms. Mechanisms of pathogen removal within these filters include adsorption to soil and filter substratum, preda-

tion (noting issues identified for zooplankton filtering above), sedimentation, physical filtering of larger organisms and inactivation due to environmental exposure (pH, temperature, sunlight [only for the hydro-botanic filter]). Both filters have been estimated to achieve 10% reduction of *E. coli* (Bruns and Peppler 2019).

Limited data is available in the literature specifically for hydro-botanic and submerge filters however a range of removal performances for microorganisms have been reported for wastewater systems using subsurface wetlands, rock filters and reed beds (reviewed by Verbyla (2015)). Bacterial reductions are in the range of 1.25 – 2.5 Log₁₀; viruses 0.5 to 2 Log₁₀ and parasitic protozoa between 0.4 and 3 Log₁₀ (Adhikari et al., 2013; Bastos et al., 2010; Garcia et al., 2010; Gerba et al., 1999; Jackson and Jackson 2008; Karim et al., 2004; Reinoso et al., 2008; Stevik et al., 2004; Vidales-Contreras et al., 2012; Vidales et al., 2003; Vymazal 2005); however noting that a key driver of removal is often the retention time within the filter, and retention times in these studies were typically in the order of days rather than hours proposed for the NSP. While the design only assumes 10% removal for each of these

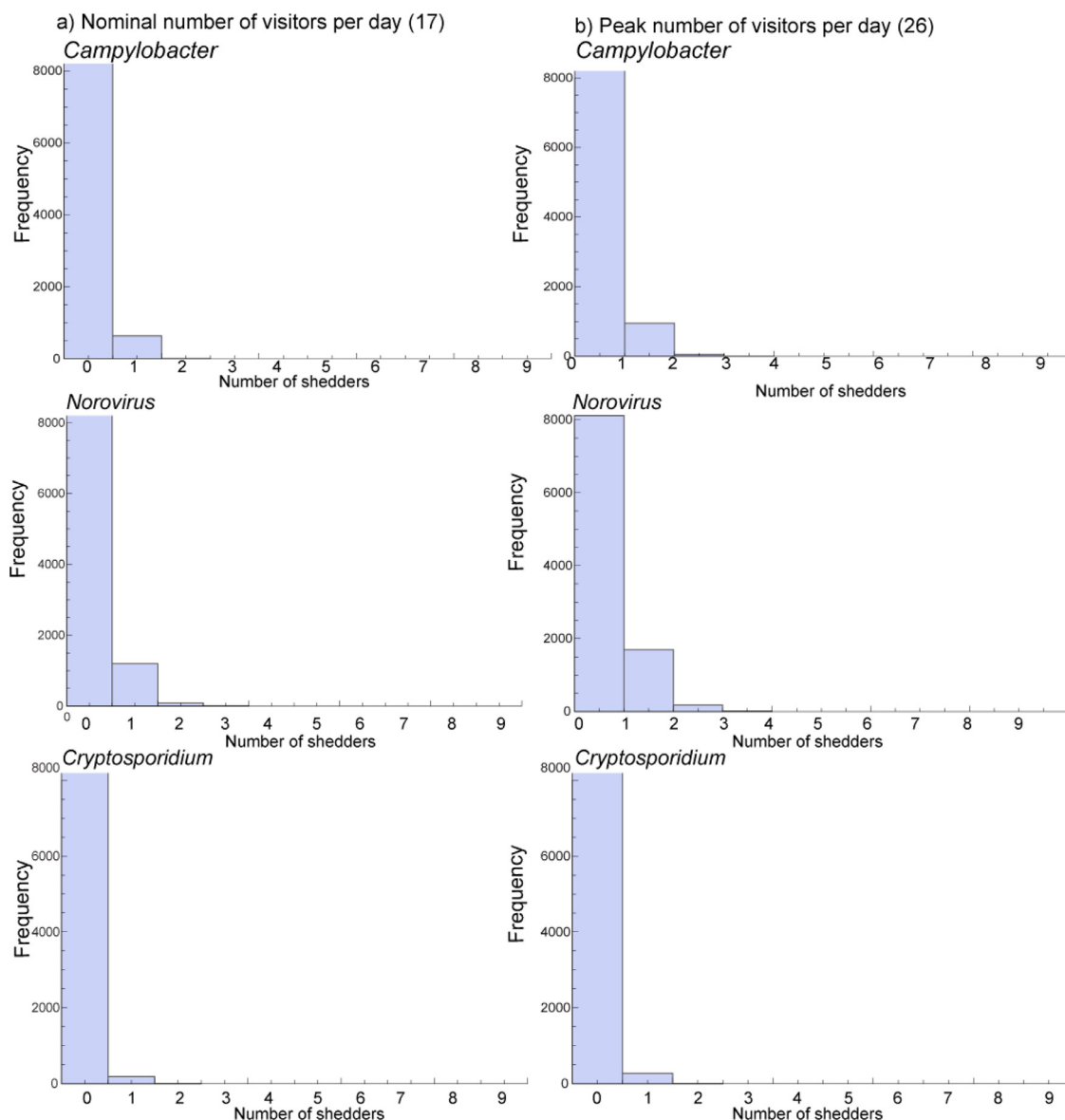


Fig. 4. Frequency histogram of the number of shedders from Monte Carlo sample (10,000 iterations) for the kiddie pool given a) nominal number of visitors and b) peak number of visitors.

filters, it seems reasonable to assume that at least 1 Log_{10} would be achieved for bacteria and protozoa, with a lower value of 0.5 Log_{10} for viruses.

Neptune filter: The Neptune filter system has a gravel medium depth of 2.00 to 2.20 m; and a retention time of 20 min was assumed by designers to have a higher *E. coli* removal efficacy of 90% in comparison to the hydro-botanic and submerse filters. Challenge studies with coliphage (assumed to be a reasonable human enteric virus surrogate to mimic their) on similar filters has shown between 95 and 99% reduction (Bruns and Peppler 2019).

UV disinfection: UV disinfection is considered as an option in the design of the treatment system. The most extensive meta-analysis review of data relating to pathogen sensitivity to UV has is by Hijnen et al. (Hijnen et al., 2006). Inactivation of reference pathogens can be estimated from the models reported in this review for the expected UV doses at the NSP system of 25 $\text{MJ}\cdot\text{cm}^{-2}$ and 40 $\text{MJ}\cdot\text{cm}^{-2}$. For bacteria and protozoa, estimated reductions were similar between reference pathogens of the same group (i.e. *E. coli* O157:H7 was similar to *Campylobacter*;

and *Cryptosporidium* was similar to *Giardia*), however for viruses, there was a strong difference between Adenoviruses compared to other enteric virus types (Ye et al., 2018). While the values for *Norovirus* inactivation are based on the calicivirus surrogate data from Hijnen et al. (2006), it is noted that if human adenovirus results were used, the Log_{10} would be 0.5, rather than 2.6.

Given a starting level of contamination of 500 *Campylobacter* per L; 10 000 *Norovirus* per L and 10 *Cryptosporidium* oocysts per L; the flow path model was run to estimate the decrease in concentration over time, and the subsequent Log_{10} reduction in the main pool and the kiddie pool, at 1 min time steps for 24 h. The model was run with and without the proposed UV disinfection units.

Given the importance of system water turnover time through treatment on the overall estimated pathogen reduction, estimated Log_{10} reductions were evaluated for turnover periods of 1, 2, 4, 6 and 8 h. Noting that current design turnover of the main pool was specified at 11 and 7.4 h for standard and maximum flow rates respectively; and in the kiddie pool, 4 and 2.7 h respectively.

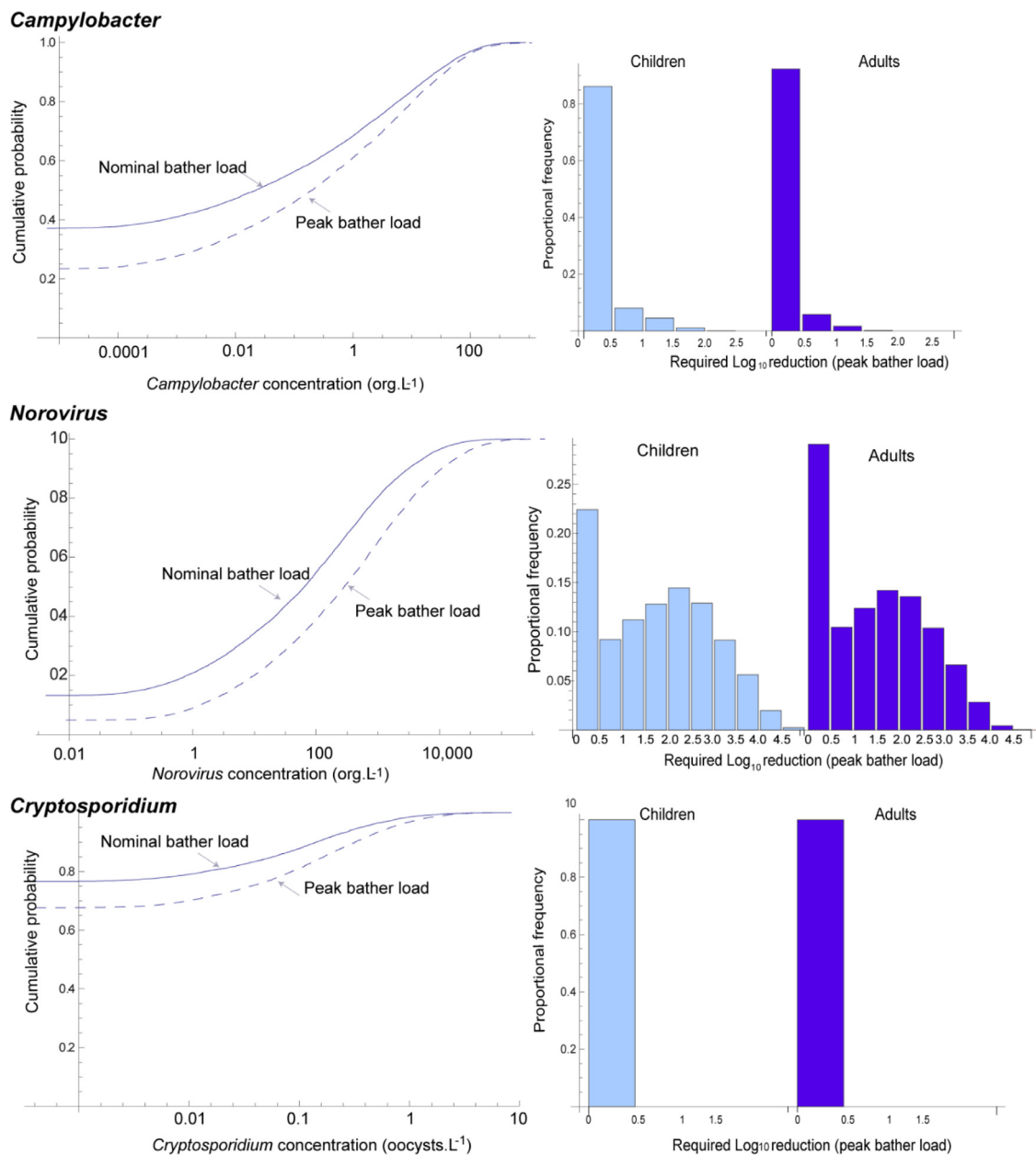


Fig. 5. Modelled concentration of reference pathogens, and Log₁₀ reduction required to achieve benchmark risk (35 illnesses per 1000) for the main pool.

2.5. Event scenarios

In addition to regular contamination, the response of the system to a larger scale faecal release event was investigated. The flow model was applied to assess the response to an accidental release by an ill swimmer. The fate of a large pathogen release (faeces or vomitus) 10^{10} *Campylobacter*; 10^{12} *Norovirus* and 10^8 *Cryptosporidium* oocysts occurring in either the main pool, kiddie pool or at the floor nozzles was modelled in all three locations over 24 h.

3. Results

3.1. Normal operation: estimated distribution of pathogen contamination

The Monte Carlo sample of the number of shedders visiting the pool per day is illustrated for the main pool and the kiddie pool in Figs. 3 & 4, respectively. The number of shed-

ders was driven by the estimated point prevalence of each reference pathogen in the population (Table 1), with the assumption of random variation following the binomial distribution used. With the highest point prevalence of *Norovirus*, 0.8%, resulted in the highest estimated number of shedders, reaching up to nine at high visitor loads. The lowest point prevalence reference pathogen used (*Cryptosporidium*, at 0.11%) led to a high likelihood of zero shedders. The resulting distribution of estimated pathogen concentration in the main pool and kiddie pool; together with the required Log₁₀ reduction to achieve the benchmark risk level is summarised in Table 5; and illustrated in Figs. 5 and 6. The highest loading and removal requirements were for enteric viruses, ranging up to 4.5 Log₁₀. The risk from *Cryptosporidium* (under baseline shedding conditions) was estimated to be extremely low. The sensitivity of the estimated pathogen loadings to the model input variables is illustrated with the Spearman Rank Correlation coefficient in Fig. 7. When prevalence was low, the estimated concentration was primarily driven by the number

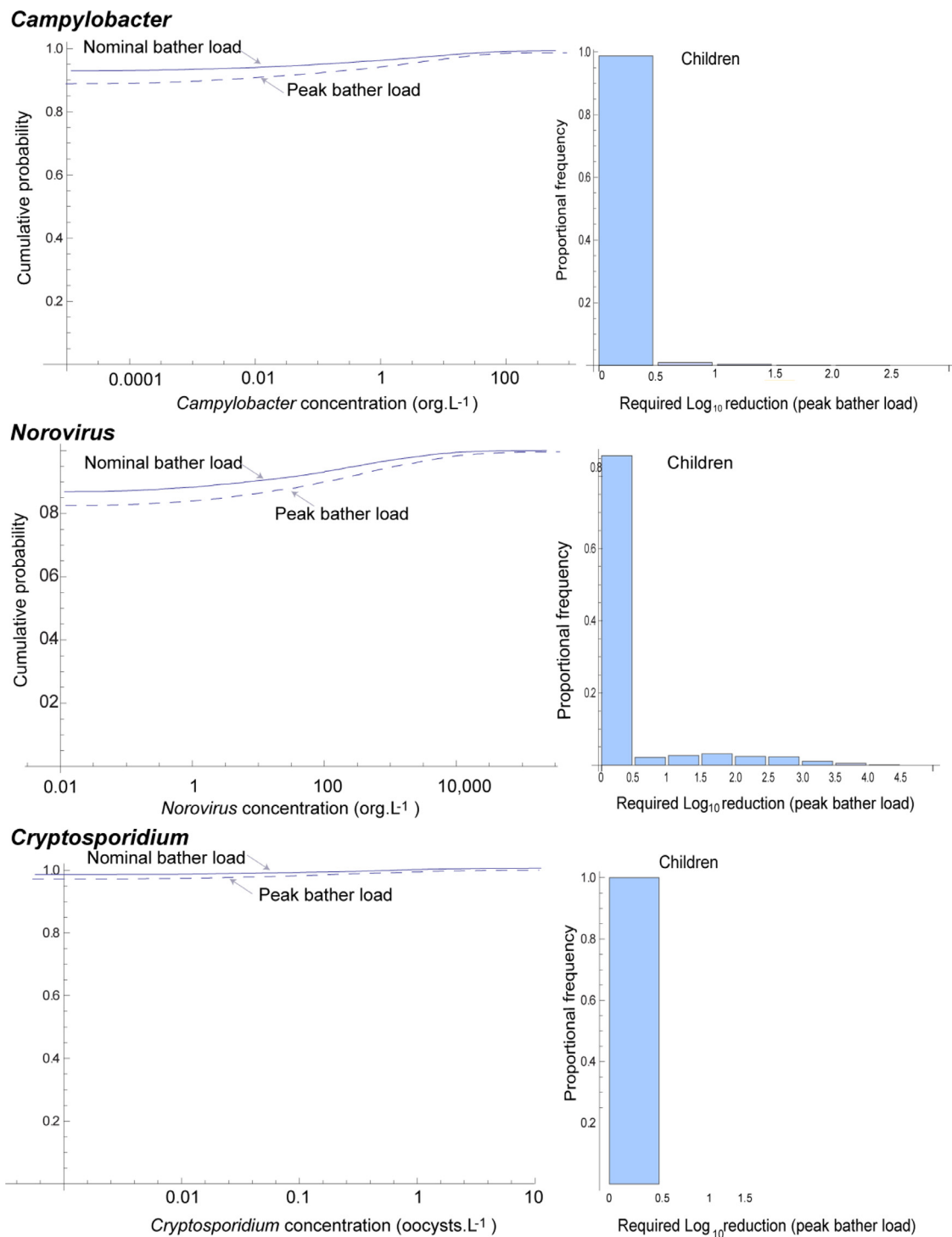


Fig. 6. Modelled concentration of reference pathogens, and Log₁₀ reduction required to achieve benchmark risk for the kiddie pool.

of shedders present in the pool. As the prevalence of infection increased (for example from *Cryptosporidium* to *Norovirus*), and hence the likelihood of one or more shedders increased, the modelled concentration was then driven by the pathogen density in faeces.

3.2. Pathogen removal capacity of the treatment system

The estimated decrease in average pathogen concentration over time, and subsequent Log₁₀ reduction in the main swimming pool is illustrated for *Norovirus* in Fig. 8 (*Campylobacter* and *Cryp-*

tosporidium are illustrated in Figures S.2 and S.3). The lower 2.5% and upper 97.5% quantiles of the Monte Carlo simulation are illustrated with dashed lines around the solid median line. In all cases, the maximum Log₁₀ reduction that could be achieved over a 24 h period in the main pool, both with and without the application of UV disinfection, was around 1 Log₁₀. In each case the achievable Log₁₀ reduction was limited by the flow rate through the external filtration/treatment system. The dashed lines (95% quantile interval of the Monte Carlo sample) represent the influence that the uncertainty in assumed pathogen removal performance by treat-

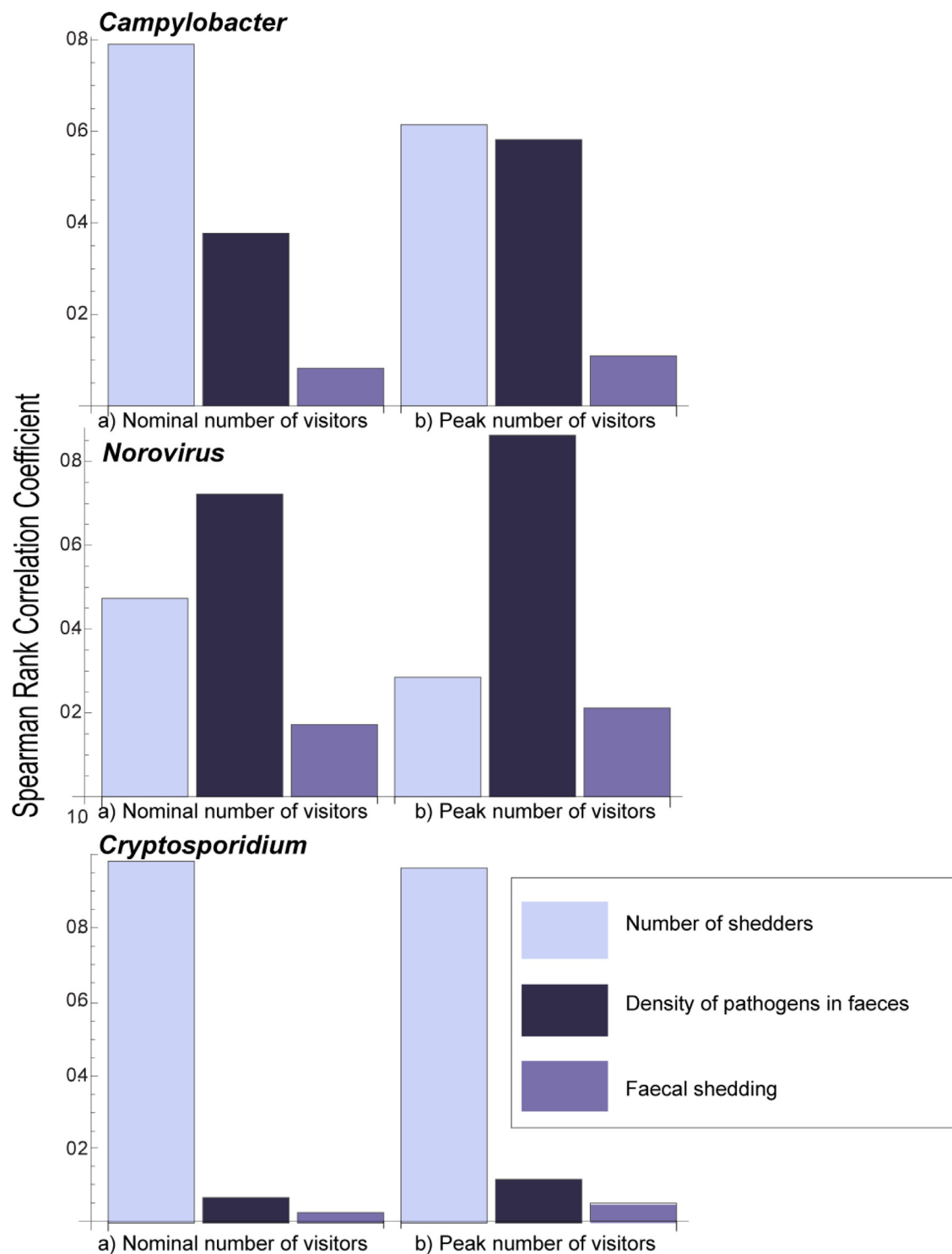


Fig. 7. Spearman rank correlation coefficient for model input samples versus estimated pathogen concentration under nominal and peak visitor numbers.

ment had on the overall estimated reduction in pathogen concentration, and hence the achieved Log_{10} reduction. This uncertainty was highest for viruses, since a low reduction was assumed across treatment barriers, and the plausible range included in the stochastic analysis was broad. However, the addition of UV disinfection eliminated the impact of this uncertainty on estimated pathogen reduction in all cases, clearly demonstrating the limiting factor of system return flow rate and pool water dilution. For the Kiddie pool, a higher overall Log_{10} reduction could be achieved within the

kiddie pool, in comparison to the main pool, due to the decrease in turnover time (4 h in comparison to 11 h for the main pool) for the system flow rates (illustrated in Figures S.4, S.5 and S.6). The maximum achievable reduction in concentration with UV disinfection was more than 3.5 Log_{10} for all pathogens (with the maximum flow rate) over 24 h.

The relationship between turnover time and achievable Log_{10} reduction for each reference pathogen is illustrated in Fig. 9. Reducing the turnover time increases the achievable Log_{10} reduction.

Table 5

Summary quantiles of Monte Carlo sample of estimated pathogen concentration and required pathogen removal to achieve benchmark* under routine bathing conditions.

	Estimated Reference			Required Log ₁₀ reduction to achieve safety*					
	Pathogen concentration (org.L ⁻¹)			Adults			Children		
	Percentile			Percentile			Percentile		
	50	95	99	50	95	99	50	95	99
MAIN POOL									
<i>Campylobacter</i>									
Nominal bathers	0.031	71	230	0	0.34	0.87	0	0.91	1.5
Peak bathers	0.23	92	290	0	0.51	1.0	0	1.1	1.6
<i>Norovirus</i>									
Nominal bathers	58	7400	29,000	0.9	3.0	3.5	1.5	3.6	4.1
Peak bathers	120	12,000	42,000	1.2	3.2	3.7	1.8	3.7	4.3
<i>Cryptosporidium</i>									
Nominal bathers	0	0.41	1.5	0	0	0	0	0	0
Peak bathers	0	0.58	1.8	0	0	0	0	0	0
KIDDIE POOL									
<i>Campylobacter</i>									
Nominal bathers	0	0.30	320				0	0	1.6
Peak bathers	0	9.4	450				0	0.14	1.8
<i>Norovirus</i>									
Nominal bathers	0	2400	38,000				0	3.0	4.3
Peak bathers	0	4500	60,000				0	3.5	4.5
<i>Cryptosporidium</i>									
Nominal bathers	0	0	1.2				0	0	0
Peak bathers	0	0	2.8				0	0	0

* benchmark of 35 illnesses per 1000 swimming events, relying on assumptions described in the text.

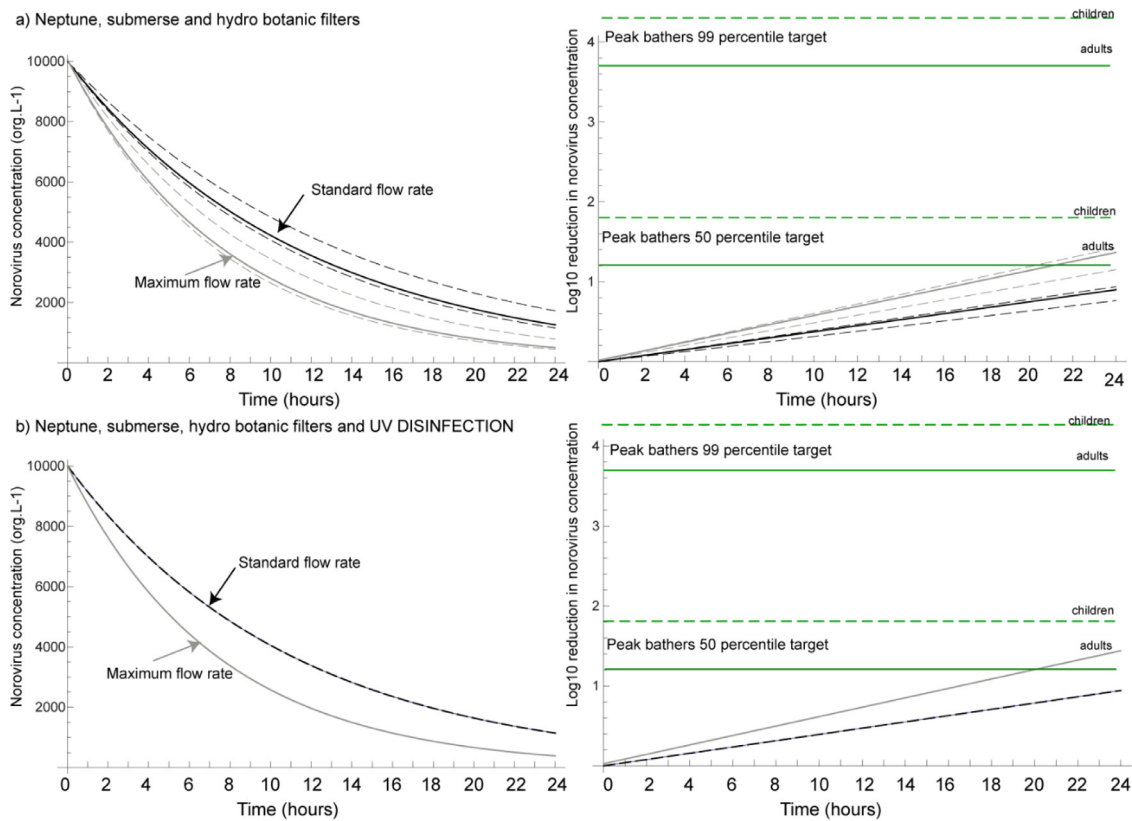


Fig. 8. Median (solid lines) and 95% quantile interval (dashed lines) of norovirus concentration and Log₁₀ reduction in main pool over time with a) Neptune, submerse and hydro botanic filters and b) with filters and UV disinfection assuming standard flow rate (black lines) and maximum flow rate (grey lines). Green lines indicate benchmark targets.

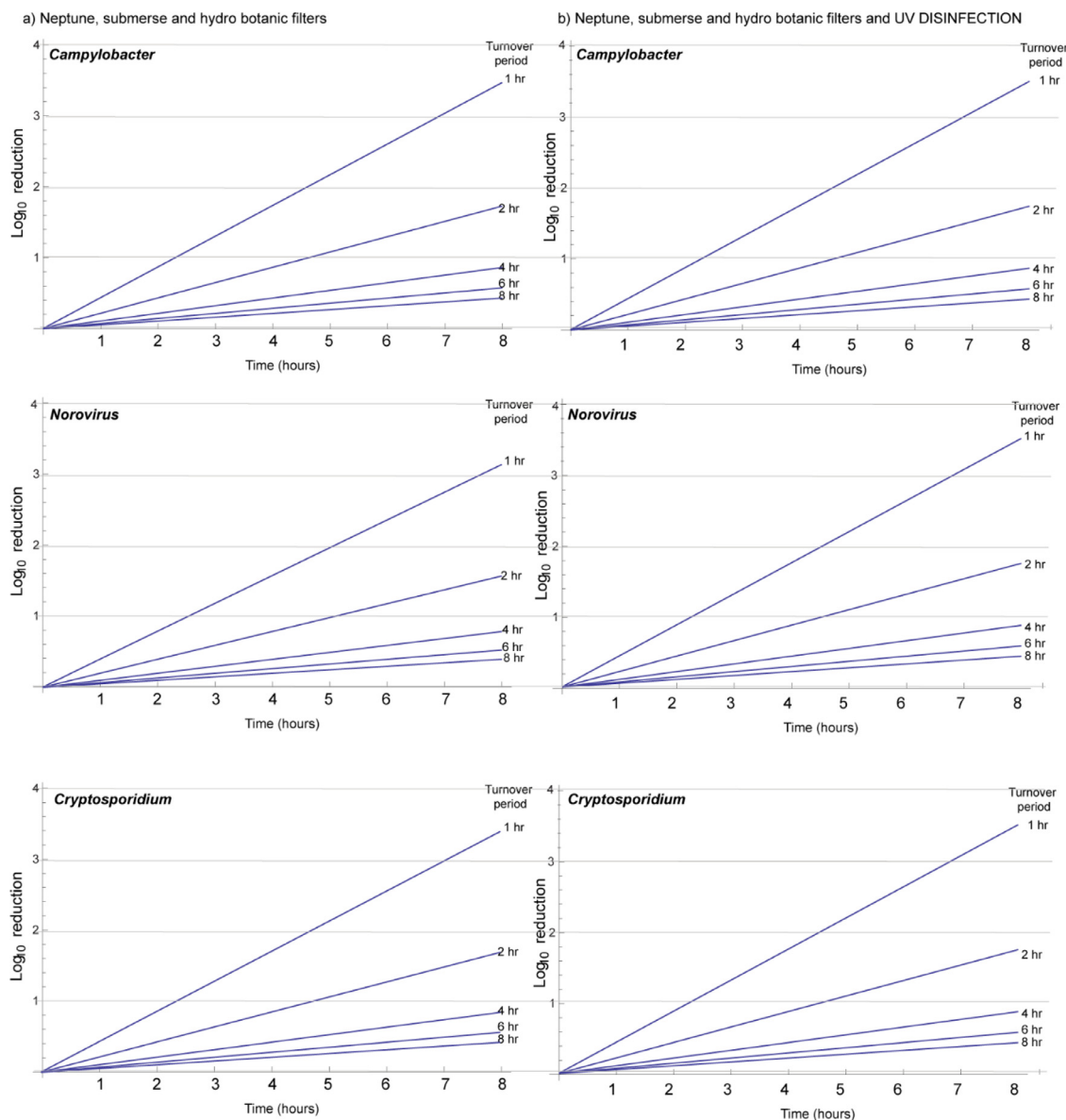


Fig. 9. Log₁₀ reduction versus time for turnover times from 1 to 8 h for: a) Neptune, submerge and hydro botanic filters and b) Neptune, submerge and hydro botanic filters with UV disinfection.

Nevertheless, achieving greater than 3 Log₁₀ reduction in concentration over 8 h, regardless of treatment barriers, would require an hourly turnover time.

3.3 Event analysis

Aside from the unintentional faecal wash-off assumed by normal bathing, accidental faecal releases (AFR) or vomitus releases can and do occur in public swimming pools. This situation was modelled as an event that could occur in one of three locations: the main pool, the kiddie pool or the floor nozzles (Fig. 2). The full results for event analysis are included in the Supplementary Material. Highest risks and hence greatest removal performance was required for *Norovirus*, followed by *Campylobacter*. Even under the modelled event scenarios, the risks from *Cryptosporidium* were calculated to be low. When the release occurred within the kiddie pool or the main pool, the recovery of the system was slow within that pool. UV disinfection did not improve this recovery period. Nevertheless, UV disinfection reduced or prevented the risks being carried across the entire system. For example for *Norovirus* risks following an event in the kiddie pool (Figure S.8), UV disinfection reduced the peak reduction requirements in the main pool from

over 4 Log₁₀ to below 2 Log₁₀, and the risk at the floor nozzles was reduced from requiring around 5 Log₁₀ to just over 2 Log₁₀. For *Campylobacter*, for a pathogen release in the main pool (Event 3), the inclusion of UV disinfection totally mitigated the risk in the kiddie pool or at the floor nozzle (Figure S.13).

4. Discussion

It is well established that the prime source of pathogen risks in artificial swimming pools are the swimmers, who unintentionally or accidentally (due to illness) shed pathogens into the water column. Hence, we estimated the likely level of pathogen contamination of the Edmonton natural swimming pool under normal operation and during potential event situations to explore how events and which pathogen groups may drive risk. Therefore, risks were summed on a Log scale and the combined risk of illness was driven by the highest risk group (enteric viruses).

The approach applied for normal operation was based on the number of visitors to the pool and relied on assumptions regarding the incidence of infection in Alberta, and estimates of faecal and

pathogen shedding from the literature. The estimated concentration was variable and uncertain, given the extreme ranges of values reported in the literature (for example shedding of noroviruses was estimated to range from 10^1 to 10^7 copies.g⁻¹). Nevertheless, some important observations are made. Firstly, given that not all individuals are expected to shed pathogens, only those who are infected with low prevalence pathogens, so the chance of no shedders present was relatively high. For our simulations under nominal visitor loading, there was more than 75% chance that no one would be shedding *Cryptosporidium* on any given day. These results highlight that the enteric pathogen risks can be transient and variable dependant upon the number of shedders present.

Furthermore, our prevalence calculations were likely to be an overestimate given that we assumed that all people would be equally likely to attend the pool, regardless of their health status or whether they were recovering from an infection. It is unlikely that an individual who was actively ill, would desire to attend the NSP, yet asymptomatic carriers would. In the absence of quantitative estimates on this, and recognizing that asymptomatic and extended shedding of pathogens (over weeks post illness) has been widely reported in the literature, it seemed wise to be cautious with the prevalence estimates and include all cases. It is important to note that the variability in the number of shedders is a critical driver of the pathogen risk, and additional attention may be needed during periods of known higher community infection rates (e.g. during an outbreak). This points to the importance of public education programs that encourage those who are or who have been unwell, to avoid swimming (as signposted at the Edmonton NSP). A message perhaps made easier given the COVID-19 pandemic.

When shedders were present and engaged in water activities the pathogen concentration and hence treatment requirements to maintain safety of other swimmers was estimated. Risk from *Cryptosporidium* were consistently low in the calculations, however a reduction of up to 2 Log₁₀ and 4 Log₁₀ were estimated to be required within the main pool for *Campylobacter* and *Norovirus* respectively. Similar values applied to the kiddie pool when shedders were present. These values were calculated for complete mixing, and hence do not account for variability (e.g. clumping) in the pathogen concentration within the pool itself at any given time. Proximity to an infected shedder may substantially increase the momentary risks. While this is potentially concerning, it is the case with all recreational swimming activities where one is in close proximity to others in waters with no residual disinfectant; what is referred to as the voluntary risk by individuals made aware of this risk when bathing with other people.

It was considered relevant to evaluate how well the proposed designed system could perform at various likely daily pathogen loads over each 8 or 12 h periods of use. This was assessed in the simplest way using a box flow model with Log₁₀ reductions applied for each treatment barrier. All the simulations presented in this study show that the key limitation to removal performance of the system was the flow rate through the external treatment barriers. The maximum that could be achieved in a 24 h period in the main pool with normal flow rates, regardless of treatment performance, was around 1 Log₁₀ for all pathogens.

The Alberta Health swimming pool standards (Alberta Health 2014) stipulate that a traditional chlorinated swimming pool constructed after November 2006 must have a turnover time of four hours. While the turnover time for a recirculating water spray park is lower at two hours, the standards also state that if 'a wading pool or recirculating water spray park is connected to a swimming pool, the turnover time for the swimming pool shall apply to the wading pool or water spray park'. Under the assumptions of the presented model, a four hour turnover time would achieve less than 1 Log₁₀ reduction for all reference pathogens. Recently the provincial agency undertaking recreational water quality testing

(ProvLab) moved to qPCR testing for *Enterococcus* spp. to indicate potential faecal contamination along with molecular testing for a human faecal marker (HF183) to detect faecal loadings, which are used on a daily basis at the Edmonton NSP to inform management.

Given that the full-scale efficiency of the external filtration system is limited by turnover time, lack of knowledge regarding the impact of zooplankton grazing on viable pathogen numbers in the water column is a critical data gap in assessing the safety of the system. Several studies have highlighted that pathogens are internalized by zooplankton in freshwater systems (Burnet et al., 2017; Connelly et al., 2007; Hahn and Höfle 2001). The question however remains as to whether internalization leads to permanent removal/inactivation of the pathogen, or whether pathogens are actually protected and their survival enhanced by internalization (Bichai et al., 2014; Bichai et al., 2008; Neogi et al., 2014). Therefore, not only is an approach for quantifying the internalization rate of pathogens by zooplankton under full scale conditions needed, but also an understanding of the ultimate fate of internalized pathogens and what health risk they may pose to bathers.

In addition, other internal inactivation processes including predation (considering now the entire microbiome/biofilm rather than zooplankton alone) and sunlight disinfection may be important health protection mechanisms. Unfortunately, the role of the natural microbiome in inactivation of pathogens is poorly understood, and at this stage not quantifiable. Investigation of the microbial inactivation within the site-specific water matrix of the NSP would be of great value.

While data does exist on sunlight inactivation of pathogens in fresh water (Bolton et al., 2010; Dahl et al., 2017; Davies-Colley et al., 1999; Fujioka and Yoneyama 2002; Mendez-Hermida et al., 2005; Silverman et al., 2015; Sinton et al., 2007; Sinton et al., 2002), the relevance of these data to the NSP situation is uncertain. Sunlight is expected to be effective at the very surface, however the extent that solar radiation can penetrate the water column is variable and likely to be affected by site specific water quality and resuspension of particulates due to bather activities. In-situ measurement of solar radiation during pool operation, at appropriate depths, would facilitate incorporation of sunlight disinfection kinetics from literature values.

To improve the safety of the system, UV disinfection was a design suggestion for the pool water return lines. However, the inclusion of UV had limited benefit on overall performance during normal operation, but could reduce the spread of infectious pathogens between pools following a faecal accident. The UV disinfection units are considered to be particularly valuable on the outflow of the floor nozzles, given the higher likelihood of faecal contamination by children in this zone, and the need to protect the larger pools from this contamination. The Log₁₀ reduction value assumed for viruses was based on calicivirus results reviewed by Hijnen et al. (2006) representative for human noroviruses (2.6 Log₁₀), but not as conservative as expected from some human adenovirus (0.5 Log₁₀). Hence the protection provided by UV disinfection may be less for some human enteric viruses than shown by these preliminary modelling results for *Norovirus*.

The predicted performance of the UV units was based on a review of published laboratory studies for a range of different pathogens (Hijnen et al., 2006). There are two important limitations linked with using this laboratory data. Firstly, the performance of UV disinfection in natural waters will be suppressed by the variable organic content of the water. Validation of the UV units would be required to ensure that the required effective UV dose is achieved using an approach similar to that recommended for drinking water treatment (USEPA 2006b). Secondly, the impact of UV disinfection on the natural biome of the water column and hence the natural elimination efficiency of the overall system is poorly understood. Disrupting the natural biome through UV disin-

fection must be undertaken with caution. Overall, in situ challenge testing trials are recommended once the system is operational to investigate the impact of the UV disinfection on microbial survival.

5. Conclusions

A Screening Level Risk Assessment is a valuable starting point for assessing waterborne risks from enteric pathogens, and to identify risk drivers and research needs. Even with the simplistic box flow model applied to the NSP described, the following important lessons regarding the likely performance of the system were identified:

- Enteric pathogen risks associated with natural swimming pools depend upon how many swimmers are infected. Modelling pathogen concentration needs to account for the likelihood of one or more shedders being present;
- The overall performance of the filtration system was driven by the system water turnover time, and using the 11 and 4 h designed turnover time would achieve less than 1 Log₁₀ reduction for all reference pathogens evaluated;
- Natural disinfection mechanisms for NSP are poorly understood, making reliance upon them for health protection in public pools challenging. Specific data is needed to better describe the fate of pathogens in natural waters; and
- Performance of natural filtration barriers in the NSP environment are poorly understood and challenge studies of in situ systems are recommended to reduce uncertainties in the current study.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Supplementary materials

Supplementary material associated with this article can be found, in the online version, at doi:[10.1016/j.watres.2020.116501](https://doi.org/10.1016/j.watres.2020.116501).

References

- Adhikari, U., Harrigan, T., Reinhold, D., Waldhorn, A.A., 2013. Modeling seasonal variation in bacteriophage removal in constructed wetlands using convection-dispersion equation. *Ecol. Eng.* 54, 266–272.
- Agasild, H., Nøges, T., 2005. Cladoceran and rotifer grazing on bacteria and phytoplankton in two shallow eutrophic lakes: in situ measurement with fluorescent microspheres. *J. Plankton Res.* 27 (11), 1155–1174.
- Ashbolt, N.J., 2015. Microbial contamination of drinking water and human health from community water systems. *Curr. Environ. Health Rep.* 2 (1), 95–106.
- Bambic, D., McBride, G., Miller, W., Stott, R., Wuertz, S., 2011. Quantification of pathogens and sources of microbial indicators for QMRA in recreational waters. *Water Intelligence Online* 10 9781843395430.
- Banting, G.S., Braithwaite, S., Scott, C., Kim, J., Jeon, B., Ashbolt, N., Ruecker, N., Tjemsens, L., Charest, J., Pintar, K., 2016. Evaluation of various *Campylobacter*-specific quantitative PCR (qPCR) assays for detection and enumeration of *Campylobacteraceae* in irrigation water and wastewater via a miniaturized most-probable-number-qPCR assay. *Appl. Environ. Microbiol.* 82 (15), 4743–4756.
- Barna, Z., Kádár, M., 2012. The risk of contracting infectious diseases in public swimming pools: a review. *Annali dell'Istituto Superiore di Sanità* 48 (4), 374–386.
- Bastos, R.K., Calijuri, M., Bevilacqua, P., Rios, E., Dias, E., Capelete, B., Magalhães, T., 2010. Post-treatment of UASB reactor effluent in waste stabilization ponds and in horizontal flow constructed wetlands: a comparative study in pilot scale in Southeast Brazil. *Water Sci. Technol.* 61 (4), 995.
- Bichai, F., Barbeau, B., Dullemont, Y., Hijnen, W., 2010. Role of predation by zooplankton in transport and fate of protozoan (oo) cysts in granular activated carbon filtration. *Water Res.* 44 (4), 1072–1081.
- Bichai, F., Dullemont, Y., Hijnen, W., Barbeau, B., 2014. Predation and transport of persistent pathogens in GAC and slow sand filters: a threat to drinking water safety? *Water Res.* 64, 296–308.
- Bichai, F., Payment, P., Barbeau, B., 2008. Protection of waterborne pathogens by higher organisms in drinking water: a review. *Can. J. Microbiol.* 54 (7), 509–524.
- Black, R.E., Levine, M.M., Clements, M.L., Hughes, T.P., Blaser, M.J., 1988. Experimental *Campylobacter jejuni* infection in humans. *J. Infect. Dis.* 157, 472–479.
- Blostein, J., 1991. Shigellosis from swimming in a park pond in Michigan. *Public Health Rep.* 106 (3), 317.
- Bolton, N., Cromar, N., Hallsworth, P., Fallowfield, H., 2010. A review of the factors affecting sunlight inactivation of micro-organisms in waste stabilisation ponds: preliminary results for enterococci. *Water Sci. Technol.* 61 (4), 885–890.
- Brunns, S., Peppler, C., 2019. Hygienic quality of public natural swimming pools (NSP). *Water Supply* 19 (2), 365–370.
- Burnet, J.-B., Faraj, T., Cauchie, H.-M., Joaquim-Justo, C., Servais, P., Prévost, M., Dörner, S.M., 2017. How does the cladoceran *Daphnia pulex* affect the fate of *Escherichia coli* in water? *PLoS ONE* 12 (2), e0171705.
- Casanovas-Massana, A., Blanch, A.R., 2013. Characterization of microbial populations associated with natural swimming pools. *Int. J. Hyg. Environ. Health* 216 (2), 132–137.
- Chalmers, R.M., 2012. Waterborne outbreaks of cryptosporidiosis. *Annali dell'Istituto Superiore di Sanità* 48, 429–446.
- Connelly, S.J., Wolyniak, E.A., Dieter, K.L., Williamson, C.E., Jellison, K.L., 2007. Impact of zooplankton grazing on the excystation, viability, and infectivity of the protozoan pathogens *Cryptosporidium parvum* and *Giardia lamblia*. *Appl. Environ. Microbiol.* 73 (22), 7277–7282.
- Dahl, N., Woodfield, P., Lemckert, C., Stratton, H., Roiko, A., 2017. A practical model for sunlight disinfection of a subtropical maturation pond. *Water Res.* 108, 151–159.
- Dale, K., Kirk, M., Sinclair, M., Hall, R., Leder, K., 2010. Reported waterborne outbreaks of gastrointestinal disease in Australia are predominantly associated with recreational exposure. *Aust. N Z J. Public Health* 34 (5), 527–530.
- Davies-Colley, R.J., Donnison, A.M., Speed, D.J., Ros, C.M., Nagels, J.W., 1999. Inactivation of faecal indicator micro-organisms in waste stabilisation ponds: interactions of environmental factors with sunlight. *Water Res.* 33 (5), 1220–1230.
- Dufour, A., Behymer, T., Cantu, R., Magnuson, M., Wymer, L., 2017. Ingestion of swimming pool water by recreational swimmers. *J. Water Health* 15 (3), 429–437.
- Dwight, R., Fernandezb, L., Bakerc, D., Semenzad, J., Olson, B., 2005. Estimating the economic burden from illnesses associated with recreational coastal water pollution—A case study in Orange County, California. *J. Environ. Manage.* 76, 95–103.
- Elmir, S.M., Shibata, T., Solo-Gabriele, H.M., Sinigalliano, C.D., Gidley, M.L., Miller, G., Plano, L.R., Kish, J., Withum, K., Fleming, L.E., 2009. Quantitative evaluation of enterococci and *Bacteroides* released by adults and toddlers in marine water. *Water Res.* 43 (18), 4610–4616.
- Elmir, S.M., Wright, M.E., Abdelzaher, A., Solo-Gabriele, H.M., Fleming, L.E., Miller, G., Rybolowik, M., Shih, M.-T.P., Pillai, S.P., Cooper, J.A., 2007. Quantitative evaluation of bacteria released by bathers in a marine water. *Water Res.* 41 (1), 3–10.
- EPA., U.S., 2012. Recreational Water Quality Criteria. U.S. Environmental Protection Agency, Washington, D.C.
- Eydeler, I. and Spieker, J. (2010) Keimelimination durch Zooplankton. *Archiv des Badewesens* (March), 167–175.
- Fewtrell, L., Kay, D., 2015. Recreational water and infection: a review of recent findings. *Curr. Environ. Health Rep.* 2 (1), 85–94.
- Folkens, M., Dey, R., Ashbolt, N.J., 2020. Interactions Between Human Reovirus and Free-Living Amoebae: implications for enteric virus disinfection and aquatic persistence. *Environ. Sci. Technol.*
- Fujioka, R.S., Yoneyama, B.S., 2002. Sunlight inactivation of human enteric viruses and fecal bacteria. *Water Sci. Technol.* 46 (11–12), 291–295.
- Garcia, J., Rousseau, D.P., Morato, J., Lesage, E., Matamoros, V., Bayona, J.M., 2010. Contaminant removal processes in subsurface-flow constructed wetlands: a review. *Crit. Rev. Environ. Sci. Technol.* 40 (7), 561–661.
- Gerba, C.P., 2000. Assessment of enteric pathogen shedding by bathers during recreational activity and its impact on water quality. *Quant. Microbiol.* 2, 55–68.
- Gerba, C.P., Thurston, J.A., Falabi, J.A., Watt, P.M., Karpiscak, M.M., 1999. Optimization of artificial wetland design for removal of indicator microorganisms and pathogenic protozoa. *Water Sci. Technol.* 40 (4–5), 363–368.
- Graciaa, D.S., Cope, J.R., Roberts, V.A., Cikes, B.L., Kahler, A.M., Vigar, M., Hilborn, E.D., Wade, T.J., Backer, L.C., Montgomery, S.P., 2018. Outbreaks associated with untreated recreational water—United States, 2000–2014. *Am. J. Transp.* 18 (8), 2083–2087.
- Guy, R.A., Arsenault, J., Kotchi, S.O., Gosselin-Théberge, M., Champagne, M.-J., Berthiaume, P., 2018. *Campylobacter* in recreational lake water in southern Quebec, Canada: presence, concentration, and association with precipitation and ruminant farm proximity. *J. Water Health* 16 (4), 516–529.
- Hahn, M.W., Höfle, M.G., 2001. Grazing of protozoa and its effect on populations of aquatic bacteria. *FEMS Microbiol. Ecol.* 35 (2), 113–121.
- Hamilton, K.A., Waso, M., Reyneke, B., Saiedi, N., Levine, A., Lalancette, C., Besner, M.C., Khan, W., Ahmed, W., 2018. *Cryptosporidium* and *Giardia* in wastewater and surface water environments. *J. Environ. Qual.* 47 (5), 1006–1023.
- Havelaar, A.H., van Pelt, W., Ang, C.W., Wagenaar, J.A., van Putten, J.P., Gross, U., Newell, D.G., 2009. Immunity to *Campylobacter*: its role in risk assessment and epidemiology. *Crit. Rev. Microbiol.* 35 (1), 1–22.
- Hijnen, W.A., Beerendonk, E.F., Medema, G.J., 2006. Inactivation credit of UV radiation for viruses, bacteria and protozoan (oo)cysts in water: a review. *Water Res.* 40 (1), 3–22.
- Health, Alberta, 2014. Pool Standards, Health System Accountability and Performance. Alberta Government.

- Jackson, E.F., Jackson, C.R., 2008. Viruses in wetland ecosystems. *Freshw. Biol.* 53 (6), 1214–1227.
- Karim, M.R., Manshadi, F.D., Karpiscak, M.M., Gerba, C.P., 2004. The persistence and removal of enteric pathogens in constructed wetlands. *Water Res.* 38 (7), 1831–1837.
- Landscape and Landscape Development Research Society, 2011. Recommendations for planning, construction, servicing and operating of outdoor swimming pools with biological water purification (swimming and bathing ponds). *Forschungsgesellschaft Landschaftsentwicklung Landschaftsbau e. V.*
- Littlewood, M., 2005. *Natural Swimming pools: an Inspirational Guide For Construction and Maintenance*. Schiffer Publishing, Pennsylvania.
- Mendez-Hermida, F., Castro-Hermida, J.A., Ares-Mazas, E., Kehoe, S.C., McGuigan, K.G., 2005. Effect of batch-process solar disinfection on survival of *Cryptosporidium parvum* oocysts in drinking water. *Appl. Environ. Microbiol.* 71 (3), 1653–1654.
- Messner, M.J., Berger, P., Nappier, S.P., 2014. Fractional poisson—a simple dose-response model for human norovirus. *Risk Anal.* 34 (10), 1820–1829.
- Neogi, S.B., Yamasaki, S., Alam, M., Lara, R.J., 2014. The role of wetland microinvertebrates in spreading human diseases. *Wetlands Ecol. Manage.* 22 (5), 469–491.
- Paunio, M., Pebody, R., Keskimäki, M., Kokki, M., Ruutu, P., Oinonen, S., Vuotari, V., Siitonen, A., Lahti, E., Leinikki, P., 1999. Swimming-associated outbreak of *Escherichia coli* O157 [ratio] H7. *Epidemiol. Infect.* 122 (01), 1–5.
- Pond, K., 2005. Water Recreation and disease: Plausibility of Associated infections: Acute effects, sequelae, and Mortality. World Health Organization.
- Qiu, Y., Lee, B.E., Neumann, N., Ashbolt, N., Craik, S., Maal-Bared, R., Pang, X., 2015. Assessment of human virus removal during municipal wastewater treatment in Edmonton, Canada. *J. Appl. Microbiol.* 119 (6), 1729–1739.
- Reinoso, R., Torres, L.A., Becares, E., 2008. Efficiency of natural systems for removal of bacteria and pathogenic parasites from wastewater. *Sci. Total Environ.* 395 (2–3), 80–86.
- Silverman, A.I., Nguyen, M.T., Schilling, I.E., Wenk, J., Nelson, K.L., 2015. Sunlight inactivation of viruses in open-water unit process treatment wetlands: modeling endogenous and exogenous inactivation rates. *Environ. Sci. Technol.* 49 (5), 2757–2766.
- Sinclair, R., Jones, E., Gerba, C., 2009. Viruses in recreational water-borne disease outbreaks: a review. *J. Appl. Microbiol.* 107 (6), 1769–1780.
- Sinton, L., Hall, C., Braithwaite, R., 2007. Sunlight inactivation of *Campylobacter jejuni* and *Salmonella enterica*, compared with *Escherichia coli*, in seawater and river water. *J. Water Health* 5 (3), 357–365.
- Sinton, L.W., Hall, C.H., Lynch, P.A., Davies-Colley, R.J., 2002. Sunlight inactivation of fecal indicator bacteria and bacteriophages from waste stabilization pond effluent in fresh and saline waters. *Appl. Environ. Microbiol.* 68 (3), 1122–1131.
- Stehr-Green, J.K., McCaig, L., Remsen, H.M., Rains, C.S., Fox, M., Juranek, D.D., 1987. Shedding of oocysts in immunocompetent individuals infected with *Cryptosporidium*. *Am. J. Trop. Med. Hygiene* 36 (2), 338–342.
- Stevik, T.K., Aa, K., Ausland, G., Hanssen, J.F., 2004. Retention and removal of pathogenic bacteria in wastewater percolating through porous media: a review. *Water Res.* 38 (7), 1355–1367.
- Stott, R., May, E., Ramirez, E., Warren, A., 2003. Predation of *Cryptosporidium* oocysts by protozoa and rotifers: implications for water quality and public health. *Water Sci. Technol.* 47 (3), 77–83.
- Suppes, L.M., Canales, R.A., Gerba, C.P., Reynolds, K.A., 2016. *Cryptosporidium* risk from swimming pool exposures. *Int. J. Hyg. Environ. Health* 219 (8), 915–919.
- Tam, C.C., Rodrigues, L.C., Viviani, L., Dodds, J.P., Evans, M.R., Hunter, P.R., Gray, J.J., Letley, L.H., Rait, G., Tompkins, D.S., 2012. Longitudinal study of infectious intestinal disease in the UK (IID2 study): incidence in the community and presenting to general practice. *Gut* 61 (1), 69–77.
- Tang, K.W., Dziallas, C., Grossart, H.P., 2011. Zooplankton and aggregates as refuge for aquatic bacteria: protection from UV, heat and ozone stresses used for water treatment. *Environ. Microbiol.* 13 (2), 378–390.
- Teunis, P., van den Brandhof, W., Nauta, M., Wagenaar, J., van den Kerkhof, H. and van Pelt, W. (2005) A reconsideration of *Campylobacter* dose-response relation *Epidemiology and infection* (133), 583–592.
- Teunis, P.F.M., Chappell, C.L., Okhuysen, P.C., 2002. *Cryptosporidium* dose response studies: variation between isolates. *Risk Anal.* 22 (1), 175–183.
- Teunis, P.F.M., Moe, C.L., Liu, P., Miller, S.E., Lindesmith, L., Barie, R.S., Pendu, J.L., Calderon, R.L., 2008. Norwalk Virus: how Infectious is It? *J. Med. Virol.* 80, 1468–1476.
- Thomas, M.K., Murray, R., Flockhart, L., Pintar, K., Pollari, F., Fazil, A., Nesbitt, A., Marshall, B., 2013. Estimates of the burden of foodborne illness in Canada for 30 specified pathogens and unspecified agents, circa 2006. *Foodborne Pathog. Dis.* 10 (7), 639–648.
- Trout, J., Walsh, E., Fayer, R., 2002. Rotifers ingest *Giardia* cysts. *J. Parasitol.* 88 (5), 1038–1040.
- Tu, E.T., Bull, R.A., Kim, M.J., McIver, C.J., Heron, L., Rawlinson, W.D., White, P.A., 2008. Norovirus excretion in an aged-care setting. *J. Clin. Microbiol.* 46 (6), 2119–2121.
- USEPA, 2006a. Economic Analysis For the Final Long Term 2 Enhanced Surface Water Treatment Rule. US EPA, Washington DC.
- USEPA, 2006b. Ultraviolet Disinfection Guidance Manual For the Final Long Term 2 Enhanced Surface Water Treatment Rule. United States Environmental Protection Agency, Office of Water (4601), Washington, DCUSA.
- Verbyla, M.E. (2015) Pathogen removal in natural wastewater treatment and resource recovery systems: solutions for small cities in an urbanizing world.
- Vidales-Contreras, J.A., Gerba, C.P., Karpiscak, M.M., Valdez-Cepeda, R.D., Hernandez-Escareno, J.J., 2012. Transport and removal of coliphage PRD1 in constructed wetlands. *J. Environ. Sci. Health, Part A* 47 (1), 142–148.
- Vidales, J.A., Gerba, C.P., Karpiscak, M.M., 2003. Virus removal from wastewater in a multispecies subsurface-flow constructed wetland. *Water Environ. Res.* 75 (3), 238–245.
- Vose, D., 2008. Risk analysis: a Quantitative Guide. John Wiley & Sons.
- Vymazal, J., 2005. Removal of enteric bacteria in constructed treatment wetlands with emergent macrophytes: a review. *J. Environ. Sci. Health* 40 (6–7), 1355–1367.
- WHO, 2016. Quantitative Microbial Risk Assessment for Water Safety Management. World Health Organization, Geneva.
- Ye, Y., Chang, P.H., Hartert, J., Wigginton, K.R., 2018. Reactivity of enveloped virus genome, proteins, and lipids with free chlorine and UV254. *Environ. Sci. Technol.* 52 (14), 7698–7708.