

Comparing the microbial risks associated with household drinking water supplies used in peri-urban communities of Phnom Penh, Cambodia

K. Thomas, E. McBean, A. Shantz and H. M. Murphy

ABSTRACT

Most Cambodians lack access to a safe source of drinking water. Piped distribution systems are typically limited to major urban centers in Cambodia, and the remaining population relies on a variety of surface, rain, and groundwater sources. This study examines the household water supplies available to Phnom Penh's resettled peri-urban residents through a case-study approach of two communities. A quantitative microbial risk assessment is performed to assess the level of diarrheal disease risk faced by community members due to microbial contamination of drinking water. Risk levels found in this study exceed those associated with households consuming piped water. Filtered and boiled rain and tank water stored in a kettle, bucket/cooler, bucket with spigot or a 500 mL bottle were found to provide risk levels within one order-of-magnitude to the piped water available in Phnom Penh. Two primary concerns identified are the negation of the risk reductions gained by boiling due to prevailing poor storage practices and the use of highly contaminated source water.

Key words | Cambodia, diarrheal disease, ETEC, household water supplies, QMRA

K. Thomas
E. McBean
H. M. Murphy (corresponding author)
School of Engineering,
University of Guelph, Guelph,
Ontario,
Canada N1G 2W1
E-mail: murph.hm@gmail.com

A. Shantz
Resource Development International Cambodia
(RDIC),
Royal Brick Road,
Kandal,
Cambodia

INTRODUCTION

For the inhabitants of the developing world, access to safe drinking water can be a daily struggle. In particular, water-related health problems persist as a result of microbiological contamination of water sources (WHO 2006). As a result, drinking-water is '... still a major contributor to the community burden of enteric disease because available water sources are faecally contaminated and untreated, inadequately treated, or have become contaminated during collection, handling, storage and use' (Havelaar & Melse 2003). This description applies to Cambodia, where an estimated 38% of the population lacks access to improved drinking water sources (UNICEF/WHO 2012); the challenges include the elevated risk of diarrheal disease due to drinking water consumption for residents in the peri-urban areas of Phnom Penh.

Risk assessment and management has become an increasingly important tool to assess water safety (WHO

2006). A particular type of risk assessment, quantitative microbial risk assessment (QMRA), has been incorporated in the WHO's (2011) 4th edition of the *Guidelines for Drinking Water Quality*.

With rapid urbanization and increasing land values in Cambodia, there are widespread trends to displace the urban poor from central city locations to the outskirts, or peri-urban areas. This situation is widespread in Phnom Penh. Consequently, these households see a change in their access to piped water, and after resettlement often need to rely on a variety of lesser quality water sources.

This paper describes the application of a QMRA methodology to quantify diarrheal disease risk, building on previous works (e.g., Howard *et al.* 2006; Hunter *et al.* 2009) that applied QMRA to the scenarios for the developing world. The QMRA framework utilized herein applies a

Latin hypercube analysis to water quality results from two resettled peri-urban communities in Cambodia, to better understand disease potential arising from various drinking water sources and storage conditions.

MATERIALS AND METHODS

Study site selection

Two peri-urban communities were selected for assessment. The criteria used in site selection of the communities were as follows:

- had no access to piped water,
- had access to several possible water sources, and
- were resettled from central Phnom Penh.

For simplicity, these two communities will be referred to as Veal Sbov (VS) and Prey Sala (PS), respectively, although these names refer to the communes where the communities are located, rather than the specific communities themselves.

The residents of VS and PS have access to several possible water sources. The water sources available represent a downgrade in quality and access, compared to the piped water they used prior to resettlement. Summaries of the drinking water sources available in both communities are presented in [Tables 1](#) and [2](#).

Household recruitment

Household eligibility criteria for participation in this research included:

- households who stored drinking water at the household level,
- households who stated that they did not mix drinking water types within a given storage vessel, and
- the head of household and/or primary caregiver agreed to voluntarily participate.

The study participants included 46 households: 20 from VS and 26 from PS. Not all households were available on a weekly basis, and two households dropped out prior to the end of the study.

Table 1 | Summary of drinking water resources available in VS

Water type	WHO classification	Details
Rainwater	Improved	<ul style="list-style-type: none"> • Primary water source used in the rainy season • Low storage capacity
Unprotected shallow wells	Unimproved	<ul style="list-style-type: none"> • Three cement-walled and one hand-dug shallow wells are present • Commonly used, often with no form of treatment • Elevated arsenic contamination levels have been detected
Surface water	Unimproved	<ul style="list-style-type: none"> • A lake is present adjacent to the site • Frequently site of garbage and waste disposal • Not commonly used for drinking water
Bottled water	Unimproved	<ul style="list-style-type: none"> • 20 L bottles of treated water sold in the community • Origin of water is not known

Table 2 | Summary of drinking water resources available in PS

Water type	WHO classification	Details
Rainwater	Improved	<ul style="list-style-type: none"> • Primary water source during the rainy season • Most households own more than one rainwater harvesting jar
Tube well	Improved	<ul style="list-style-type: none"> • Two tube wells of unknown depth are present on the site • Residents judge water as being poor quality due to poor esthetics • Elevated iron levels
Unprotected shallow wells	Unimproved	<ul style="list-style-type: none"> • One communal concrete-lined shallow well is present • Used by only one study household
Tanker truck water	Unimproved	<ul style="list-style-type: none"> • Tanker truck delivers water to the community • Source of water is unknown • Delivery may take place to individual households directly or to a privately owned water tank in the community

Ethics approval

Free and informed consent of the participants was obtained and the study protocol was approved by the Committee for the Protection of Human Subjects – Research Ethics Board at the University of Guelph, Ontario, Canada, Protocol #09JN018, approved 7 December 2009.

Data collection and analysis

The study consisted of 11 weeks of sampling conducted over a 12-week period. Study participants were asked to provide drinking water samples, treating the sample bottle as if it was a household drinking cup. Therefore, the sample collected was the source of drinking water that would be consumed at that time in the household, whether it was boiled, filtered, or untreated. Water samples of 250 mL in volume were collected from each household at the time of each visit and used to test for *Escherichia coli* and total coliform (TC) concentrations. The samples were kept in an ice-filled cooler until delivery to the laboratory, where they were refrigerated and analyzed within 24 hours.

E. coli was selected as the indicator bacteria for use in this study, as per the recommendations of the WHO (2006). *E. coli* and TC were enumerated using the membrane filtration method (Standard Method-9222B). Differential Coliform Agar with chromogenic agent BCIG (OXOID Culture Media) was used. For quality assurance, at least two dilutions were plated in duplicate for each sample. Samples were incubated for 22 to 24 hours at 37 °C, at which point they were enumerated for TC (pink/red colonies) and *E. coli* (blue-purple colonies).

Model development

QMRA is the application of the principles of risk assessment to estimate the potential effects of an exposure to infectious microorganisms (Haas *et al.* 1999). The WHO (2006) describes QMRA as: ‘... a rapidly evolving field that systematically combines available information on exposure and dose–response to produce estimates of the disease burden associated with exposure to pathogens’.

Reference level of risk

The reference level of risk for waterborne diseases recommended by the WHO (2006) is 1×10^{-6} disability adjusted life years (DALYs) per person per year. However, the WHO (2006) also states that the reference level of risk can be adapted to local circumstances on the basis of a risk–benefit approach. For the purpose of this QMRA, the reference level of risk is based on the risk associated with the consumption of untreated piped water in the city of Phnom Penh. This reference level of risk was selected because piped water was the water supply used by the households in the present study prior to resettlement in peri-urban communities. DALYs were chosen as the measure of disease burden for two reasons: (1) it is an internationally recognized measure for describing microbial risk; and (2) it allows for the results to be compared to the WHO reference level of risk and consequently, to other international studies using the same measure.

Reference pathogens

A key element in QMRA is the selection of reference pathogens. WHO (2006) recommends the selection of representative organisms that ‘... if controlled, would ensure control of all pathogens of concern. Typically this implies inclusion of at least one bacterial pathogen, virus, and protozoan’. *E. coli* O157:H7 or enterotoxigenic *E. coli* (ETEC), *Cryptosporidium parvum* (*C. parvum*), and rotavirus may serve as reference pathogens for bacterial, protozoan, and viral pathogens, respectively (Howard *et al.* 2006; Hunter *et al.* 2009).

Both *E. coli* O157:H7 (Howard *et al.* 2006) and ETEC (Hunter *et al.* 2009) have been used as reference pathogens in QMRA in developing countries. In the present study, ETEC is used rather than *E. coli* O157:H7, because it is a ‘... far more significant pathogen in developing countries that is largely transmitted by food and water with little or no person to person spread’ (Hunter *et al.* 2009).

The water quality data collected in this study were limited to TCs and generic *E. coli*, as insufficient laboratory capacity was available to perform more sophisticated pathogen testing. The standard methods for detecting the presence of *E. coli* organisms in water samples in routine

water testing are not designed to detect pathogenic strains specifically (*Standard Methods 2005*). Ratios exist to relate generic *E. coli* to specific strains, although there is uncertainty in the use of such ratios. Published studies that document ETEC:*E. coli* ratios are available (e.g., van Lieverloo et al. 2007); however, these studies were conducted in developed country settings, which are not necessarily appropriate for the context of peri-urban Cambodia. A review of the literature was conducted by the authors to compile data from Asian tropical environments to develop a ratio of ETEC relative to generic *E. coli* that could be used for the context of Cambodia. The review identified six studies conducted in Bangladesh and India where concentrations of ETEC relative to generic *E. coli* were measured (Figure 1) (Begum et al. 2005, 2007; Alam et al. 2006; Ram et al. 2007, 2008, 2009). The mean of the ratios documented in these studies was used to estimate the ratio of ETEC:*E. coli* for the present study.

Weak correlations between TC and *C. parvum* correlations have been identified in the literature (Hsu et al. 1999; Anbazhagi et al. 2007; Wilkes et al. 2009). Nieminski et al. (2010) assessed the validity of using *E. coli* as a reference pathogen for *C. parvum* and found a poor correlation between the two parameters. While some studies do identify a correlation between these parameters, many publications ‘...report the limited

correlation between the presence and concentration of fecal indicators and the presence and concentration of waterborne pathogens. They demonstrate in particular that fecal indicator bacteria are poor surrogates for protozoa and viral pathogens’, as summarized in Dechense & Soyeux (2007). Dechense & Soyeux (2007) explain that different systems exhibit different behaviors, resulting in the links between microbial parameters being site-specific. Therefore, *C. parvum* was not included in this study due to the lack of literature to support the use of ratios for *E. coli* concentrations to estimate its presence.

There is a precedent for the application of ratios of *E. coli* to rotavirus concentrations in surface water (Mehnert & Stewien 1993; Lopez-Pila & Szewzyk 2000). However, this approach was not adopted since it is not known how applicable the available ratio is to surface waters in Phnom Penh.

Exposure assessment

Exposure assessment requires the estimation of the number of pathogenic microbes to which an individual is exposed; in this case, the only pathway considered was ingestion. Water quality and water consumption rate data were used to estimate exposure.

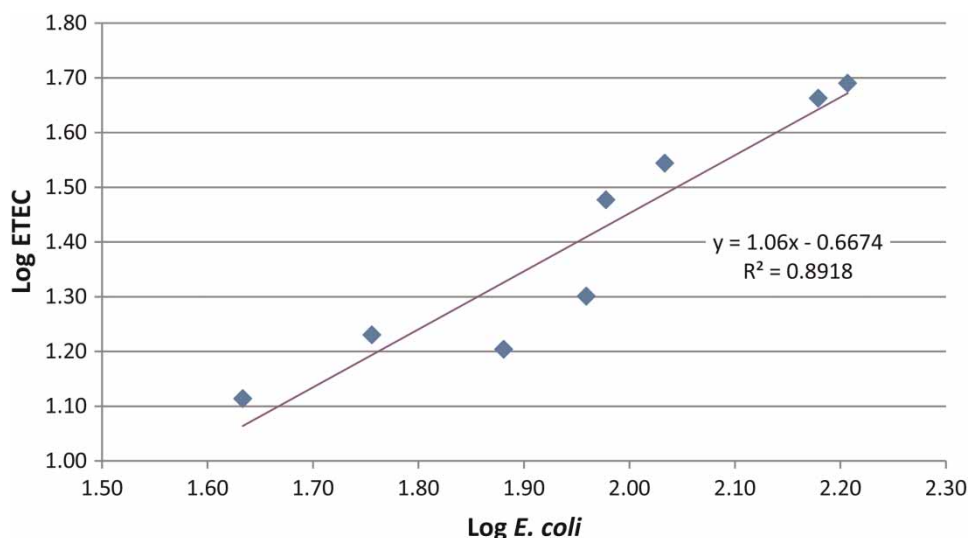


Figure 1 | Relationship between *E. coli* and ETEC (created using data from Begum et al. 2005, 2007; Alam et al. 2006; Ram et al. 2007, 2008, 2009 from data collected in India and Bangladesh).

Water quality data

Household water quality

Microbial water quality was reported as colony forming units (CFU) of generic *E. coli* per 100 mL (CFU/100 mL). Generic *E. coli* data for the different water sources were fit to lognormal distributions using maximum likelihood. Data below the detection limit (<1 CFU/100 mL) were censored in the analyses (as adopted by Haas & Eisenburg 2001). The data were fit using EasyFit 5.0 software, and the Kolmogorov–Smirnov statistic was used to determine goodness of fit to the lognormal distribution. The mean and standard deviations of these distributions were used to generate lognormal probability distributions to describe the concentrations of generic *E. coli* in the various water supplies when the supply was positive for generic *E. coli*. One key assumption was that all samples that contained *E. coli* below the detection limit (censored values) were assumed to be ‘negative’ for generic *E. coli*. Using the % of positive samples, a prevalence rate (PR) term was developed for each water source and applied when

calculating the annual probability of infection. The PR represents the likelihood that a sample is positive with generic *E. coli*.

T-test and ANOVA analyses were used to determine how to categorize and aggregate household water quality data as there were many different types of water sources/storage systems. Water samples stored in a kettle did not have statistical differences based on source water type ($p < 0.05$). Therefore, all boiled samples stored in a kettle were considered together, while samples stored in different vessels were considered as separate by source water type. A summary of the household water quality results for *E. coli* is presented in Table 3.

Piped water quality

The Phnom Penh Water Supply Authority (PPWSA) operates three water treatment plants, all of which are equipped with conventional treatment processes consisting of flocculation with alum, sedimentation, filtration, and chlorine disinfection. A free chlorine level of 0.1 mg/L is maintained

Table 3 | Summary of *E. coli* concentration results in household water supplies by water, treatment, and storage type

Household water type	n	Geometric mean (CFU/100 mL)	Number of samples \geq detection limit (1 CFU/100 mL)	Fitted lognormal parameters (values below detection level censored)	
				Mean	Std. deviation
Filtered water	27	3.19	15	1.49	2.17
Boiled water					
Kettle	47	1.96	14	1.17	2.80
Rain and tank water					
Bucket/Cooler	66	8.92	37	0.69	0.96
Bucket with spigot	24	2.75	9	1.37	2.83
Bottle (500 mL)	13	7.44	6	2.94	3.84
Bottle (20 L)	4	31.80	3	2.11 ^a	0.32 ^a
Shallow well water					
Bucket/Cooler	17	42.37	13	2.37	4.93
Untreated water					
Rainwater	118	19.49	101	1.58	3.65
Shallow well water	30	647.50	30	2.8	5.44
Bottled water	10	18.35	8	2.24	5.35
Tank water	18	22.48	16	2.11	3.53

^aToo few data to fit a lognormal distribution; lognormal distribution assumed and mean and log standard deviation reported here.

throughout the endpoints of the supply network (Chea 2010, Personal communication). Surface water is the water source for all three water treatment plants.

Daily monitoring of 38 water quality parameters is performed at the water quality laboratories located at each water treatment plant. A water quality report was provided by the PPWSA; the estimate of influent (raw water) *E. coli* concentrations used in this study was based on the results provided by the PPWSA. The PPWSA uses total and fecal coliforms as indicator organisms as summarized in Table 4. A ratio *E. coli* to fecal coliforms of 0.77 was applied to the influent TC data provided by the PPWSA to estimate the *E. coli* concentration in raw river water (after Rasmussen & Zeigler 2003).

Results from the PPWSA indicated no detected total or fecal coliforms in the treated water. An estimated treatment efficiency for conventional water treatment with chlorine disinfection of 10^5 (Howard et al. 2006) was used in the QMRA to estimate effluent water quality. Piped water was used as a 'best case' scenario, or reference level of risk, as this represented the households' previous water quality situation prior to relocation to the peri-urban communities. The piped water data set was limited to minimum, maximum, and average concentration data; therefore, for this water type, a point estimate of the mean was assigned as the concentration input of generic *E. coli* into the model.

Water consumption rate

The WHO (2006) recommends an average consumption rate of 1 L/d (WHO 2006). However, it is believed that this may greatly underestimate consumption levels in tropical developing countries (Howard & Bartram 2003; RDIC 2009). Ideally, country-specific statistical distributions for water consumption rate should be used in QMRA to reflect the

variability in consumption within the population (Mons et al. 2007). No country-specific information was available for Cambodia; therefore, in this study an average water consumption rate of 1.4 L/d and an expected range of 1.0 to 2.4, as reported in IPCS (1994) and WHO (2006) were employed. Using these parameters, a mean water consumption rate of 1.4 L/d and standard deviation of 0.233 L/d were used as input parameters for a lognormal distribution (IPCS 1994).

Dose-response

Well-dispersed pathogens in water are generally considered to be Poisson-distributed (WHO 2006). Given the variation in pathogen-host survival probability due to diversity in human response and/or pathogen competence, the beta distribution may be used to define the probability of an organism causing an infection (Haas et al. 1999). The resulting combination of these two distributions is the Beta Poisson distribution, as per Equation (1), presented in Haas et al. (1999):

$$P_I(d) = 1 - \left(1 + \frac{d}{\beta}\right)^{-\alpha} \quad (1)$$

where, $P_I(d)$ is the probability of infection, d is the mean dose of viable microorganisms, and α and β represent parameters of the microorganisms-host interaction.

The dose-response relationship may be simplified for low exposures by assuming linearity, as the probability of infection resulting from exposure to a single organism (WHO 2006). However, due to the relatively high exposures considered in this analysis, the Beta Poisson model was utilized.

Limited dose-response data for ETEC is available in the literature. The best available option is considered to be using the dose-response parameters for *E. coli* O157:H7 to represent ETEC (Hunter et al. 2009). Beta Poisson dose-response parameters for *E. coli* O157:H7 of $\alpha = 0.22$ and $\beta = 8,700$ (Powell 2000) were used in this assessment.

Risk characterization

Disease burden calculation

The calculation of the disease burden in DALYs requires two key pieces of information (Havelaar & Melse 2003):

Table 4 | PPWSA raw and calculated treated water quality

Water quality parameter	Minimum (CFU/100 mL)	Average (CFU/100 mL)	Maximum (CFU/100 mL)
Fecal coliform (raw water)	2,000	21,450	75,900
<i>E. coli</i> (raw water) ^a	1,540	16,517	58,443
<i>E. coli</i> (treated water) ^b	0.0154	0.165	0.584

^a $E. coli_{\text{raw water}} = 0.77 (FC_{\text{raw water}})$.

^b $E. coli_{\text{treated}} = E. coli_{\text{raw water}} \times (1 - 0.9999)$; (where 0.9999 = treatment effect).

1. Average duration of the adverse health response, including loss of life (D).
2. Weights for severity to the unfavorable health conditions (S), and DALY values are calculated for each negative health consequence expected to result from infection using Equation (2):

$$\text{DALY} = D \times S \quad (2)$$

DALY values are calculated for each negative health consequence expected to result from infection, and summed together, to determine the maximum disease burden.

Disease burden calculation for ETEC

The severity of response estimate for the watery diarrhea cases was based on *E. coli* O157:H7 infection by Havelaar & Melse (2003), who used a severity weighting of 0.067. The duration of watery diarrhea caused by ETEC was estimated to be between 3 and 4 days (Qadri *et al.* 2005). The proportion of symptomatic cases has been reported to range between 80 and 100% (Qadri *et al.* 2005) and 78 and 100% (Gupta *et al.* 2008).

The values of duration and severity of response for death resulting from diarrheal disease are complicated, due to the limited information available on ETEC mortality. ETEC is disregarded in many epidemiological studies largely due to difficulties in diagnosis and mortality figures are difficult to obtain as most ETEC-caused diarrheal episodes are treated in the home (Wenneras & Erling 2004). If properly treated and hydration maintained, the rate of mortality is expected to be less than 1% (Qadri *et al.* 2005) and possibly as low as <0.1% (Haas *et al.* 1999). No information was identified on the rate of mortality if adequate medical treatment was not sought. In this analysis, a uniform distribution, with a minimum value of 0.1% and a maximum likelihood value of 1%, was used to represent the rate of mortality. This distribution may be conservative, but may reflect the relatively high rate of mortality for ETEC infection in developing regions. The mortality burden was based on an average age of death of one year (Howard *et al.* 2006).

The average life expectancy at birth in Cambodia was used in the estimation of the years of life lost from premature death (the mortality fraction) and years of life impaired (the morbidity fraction). This basis is in keeping with the method of Howard *et al.* (2006) who argued that national life expectancy better reflected the impact of diseases in developing countries. The average life expectancy at birth in Cambodia is 59 for men and 64 for women (WHO 2008); the average of 61 was used herein. It is noted that the use of national life expectancy may distort the size of disease burdens toward morbidity and mortality of the very young; however, this reflects the importance of such diseases and does not cause unreasonable distortions unless one is comparing different types of data sets (Howard *et al.* 2006).

A summary of the values used in the calculation of disease burden and the results of the DALY calculations are presented in Table 5.

Using point estimates of the averages of the values presented in Table 5, an estimated maximum disease burden of 0.601 is calculated. Therefore, diarrheal disease caused by ETEC infection can be expected to result in the loss of 0.601 years of healthy life. This value represents the 'maximum disease burden' used in the risk analysis calculation. In subsequent analyses, statistical distributions are used to describe the input parameters of the maximum disease burden calculation.

Risk calculations

It is possible to perform QMRA analyses using point estimates of input parameters by combining a point estimate of exposure (typically an estimate of the average or an extreme dose) with a point estimate of dose–response

Table 5 | Disease burden estimation for ETEC infection

Outcomes	Severity	Duration	Symptomatic cases/Cases of mortality	Disease burden per cases on DALYs
Watery diarrhea	0.067	3–4 days	80–100%	5.78×10^{-4}
Death from diarrhea	1	60 years	0.1–1%	0.601

parameters to compute a point estimate of risk (WHO 2006). This approach may be useful in some developing world contexts, where there may not be access to the software packages typically used to handle statistical distributions. However, to capture the uncertainty and variability associated with the data that are incorporated into a QMRA analysis, the use of statistical distributions of input parameters is preferred. This approach, combining a characterization of the full distribution of exposure and dose–response relationships using $\times 10,000$ iterations using Latin hypercube to provide a distribution of the risk, is employed here. @Risk Version 5.7 (Palisade Corporation) was used to perform the Latin Hypercube analyses. The procedure used for calculating disease burden is described in Table 6.

Model inputs

A summary of the model inputs used in this analysis is presented in Table 6.

RESULTS AND DISCUSSION

A summary of disease burdens corresponding to different drinking water sources assuming use of a single water type for the entire year is presented in Table 7 (Figure 2). Since water use is typically seasonal in Cambodia, these estimates of annual risk are considered only for comparison across water types. Scenarios which better represent the seasonal differences in water use are described later (Table 8, Figure 3).

Table 6 | Procedure for calculating disease burden and summary of QMRA inputs (Howard et al. 2006, adapted from WHO 2006)

Variable	Description	Units	Model assumptions, values, distributions	Source
<i>E</i> _{TEC}	Concentration of pathogenic <i>E. coli</i> (E _{TEC}) in drinking water	CFU/100 mL	E _{TEC} : generic <i>E. coli</i> ; 25:1. Mean ratio, based on data from difference surface water sources in India and Bangladesh (see Figure 1)	Weekly <i>E. coli</i> data transformed into E _{TEC}
<i>C</i> _{<i>D</i>}	Drinking water quality (organisms per liter)	CFU/L	$C_D = E_{TEC}$. Lognormal distribution used for water quality data	Weekly water quality results
<i>V</i>	Volume of consumed drinking water	L	Lognormal distribution assumed. Mean = 1.4; standard deviation = 0.233	IPCS (1994)
<i>E</i>	Exposure by drinking water	CFU/L	$E = C_D \times V$	Calculated
<i>r</i>	Dose–response	Unitless	Beta-Poisson dose–response curve assumed for E _{TEC} based on <i>E. coli</i> O157:H7 $r = 1 - (1 - E/\beta)^{-\alpha}$ where $\beta = 8700$; $\alpha = 0.22$	Powell (2000); Hunter et al. (2009)
<i>P</i> _{<i>inf,d</i>}	Risk of infection per day $E \times r$	Risk inf./day	$E \times r$	Calculated
<i>PR</i>	Prevalence rate	%	% of samples that were positive (\geq detection limit of 1 CFU/100 mL) for <i>E. coli</i> indicator bacteria	Calculated
<i>P</i> _{<i>inf,y</i>}	Risk of infection per years	Risk inf./years	$P_{inf,y} = 1 - (1 - P_{inf,d})^{365} \times PR$ or approximated as $P_{inf,d} \times 365$ when $P_{inf,d} < 1$	
<i>P</i> _{<i>illjinf</i>}	Risk of diarrheal disease given infection	Risk	80–100% 78–100% Uniform distribution assumed between 0.8 and 1.0	Qadri et al. (2005) Gupta et al. (2008)
<i>P</i> _{<i>ill</i>}	Risk of diarrheal disease	Risk/year	$P_{inf,y} \times P_{ill \mid inf}$	
<i>mdb</i>	Maximum disease burden	DALY	0.601	See Table 5 for calculations
<i>fs</i>	Susceptible fraction	%	% of time water source is used for drinking (i.e., if water source is used year round $fs = 100\%$)	Determined based on weekly survey results
<i>DB</i>	Disease burden	DALY	$P_{ill} \times mdb \times fs$	

Table 7 | Summary of disease burden values for different water types (assumes use of a given water type for a 12-month period)

Water type	Disease burden in DALY				
	Mean	Lower quart.	Upper quart.	Min.	Max.
Piped water ^a	2.90×10^{-3}	2.54×10^{-3}	3.22×10^{-3}	1.47×10^{-3}	5.61×10^{-3}
Filtered water	1.20×10^{-1}	2.39×10^{-2}	2.48×10^{-1}	6.53×10^{-3}	3.34×10^{-1}
Boiled water					
Stored in kettle (all types)	4.50×10^{-2}	7.77×10^{-3}	5.86×10^{-2}	3.23×10^{-3}	1.79×10^{-1}
Rain/Tank water					
Bucket/Cooler	6.07×10^{-2}	1.51×10^{-2}	5.73×10^{-2}	6.21×10^{-3}	3.37×10^{-1}
Bucket/Cooler w/spigot	6.69×10^{-2}	1.13×10^{-2}	1.18×10^{-1}	4.46×10^{-3}	2.25×10^{-1}
Small bottle	1.62×10^{-1}	5.73×10^{-2}	2.47×10^{-1}	5.35×10^{-3}	2.77×10^{-1}
20 L bottle	3.54×10^{-1}	3.22×10^{-1}	4.03×10^{-1}	3.76×10^{-2}	4.50×10^{-1}
Shallow well					
Bucket/Cooler	1.95×10^{-1}	3.41×10^{-2}	3.87×10^{-1}	8.20×10^{-3}	4.60×10^{-1}
Untreated					
Rainwater	1.42×10^{-1}	2.29×10^{-2}	2.84×10^{-1}	9.12×10^{-3}	1.42×10^{-1}
Shallow well water	2.89×10^{-1}	5.85×10^{-2}	5.20×10^{-1}	1.15×10^{-2}	2.89×10^{-1}
Bottled water	1.87×10^{-1}	2.97×10^{-2}	3.96×10^{-1}	9.39×10^{-3}	1.87×10^{-1}
Tank water	2.32×10^{-1}	4.62×10^{-2}	4.48×10^{-1}	1.11×10^{-2}	2.32×10^{-1}

^aIt was not possible to use a statistical distribution to describe the *E. coli* concentration in raw water; therefore, this estimate is based on a point estimate of the average concentrations.

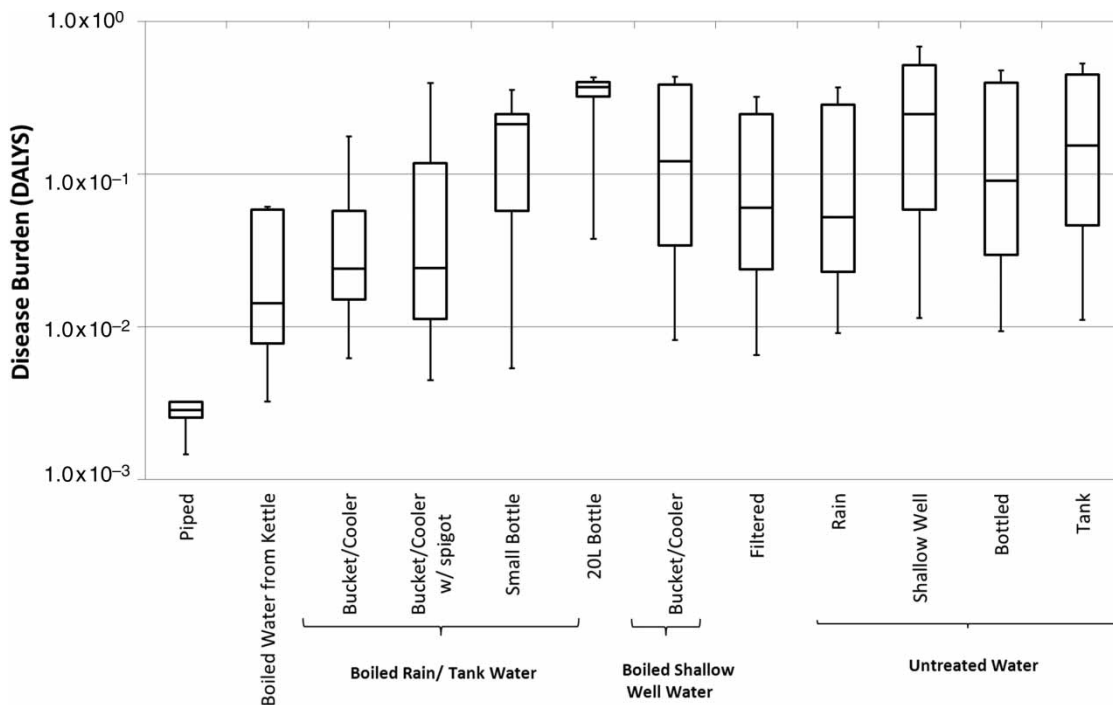
**Figure 2** | Box and whisker plot representing the median, 25th and 75th percentile, maximum, and minimum estimates of risk.

Table 8 | Summary of disease burdens for different water use scenarios

Scenario	Disease burden in DALYs					
	Mean	Lower quart.	Upper quart.	Min.	Max.	
Best case scenarios:						
1	Piped	2.90×10^{-3}	2.53×10^{-3}	3.22×10^{-3}	1.43×10^{-3}	5.49×10^{-3}
2	100% use of boiled water stored in kettle	4.50×10^{-2}	7.77×10^{-3}	5.91×10^{-2}	3.02×10^{-3}	1.79×10^{-1}
3	100% use of filtered water	1.20×10^{-1}	2.38×10^{-2}	2.47×10^{-1}	6.33×10^{-3}	3.34×10^{-1}
Typical scenarios:						
4	Rainy season: rainwater (50% boiled, bucket; 50% untreated) Dry season: tank water (50% boiled, bucket; 50% untreated)	1.24×10^{-1}	5.63×10^{-2}	1.66×10^{-1}	1.31×10^{-2}	3.98×10^{-1}
5	Rainy season: boiled rain water (40% kettle, 60% bucket) Dry season: boiled shallow well water (20% kettle, 80% bucket)	1.10×10^{-1}	4.23×10^{-2}	1.77×10^{-1}	9.66×10^{-3}	3.05×10^{-1}
6	Rainy season: untreated rain water Dry season: boiled shallow well water (20% kettle, 80% bucket)	1.50×10^{-1}	4.94×10^{-2}	2.11×10^{-1}	1.00×10^{-2}	4.08×10^{-1}
7	Rainy season: untreated rain water Dry season: untreated shallow well water	2.16×10^{-1}	8.07×10^{-2}	3.00×10^{-1}	1.47×10^{-2}	5.24×10^{-1}
Worst case scenario:						
8	100% use of untreated shallow well water year round	2.89×10^{-1}	5.87×10^{-2}	5.20×10^{-1}	1.15×10^{-2}	6.01×10^{-1}

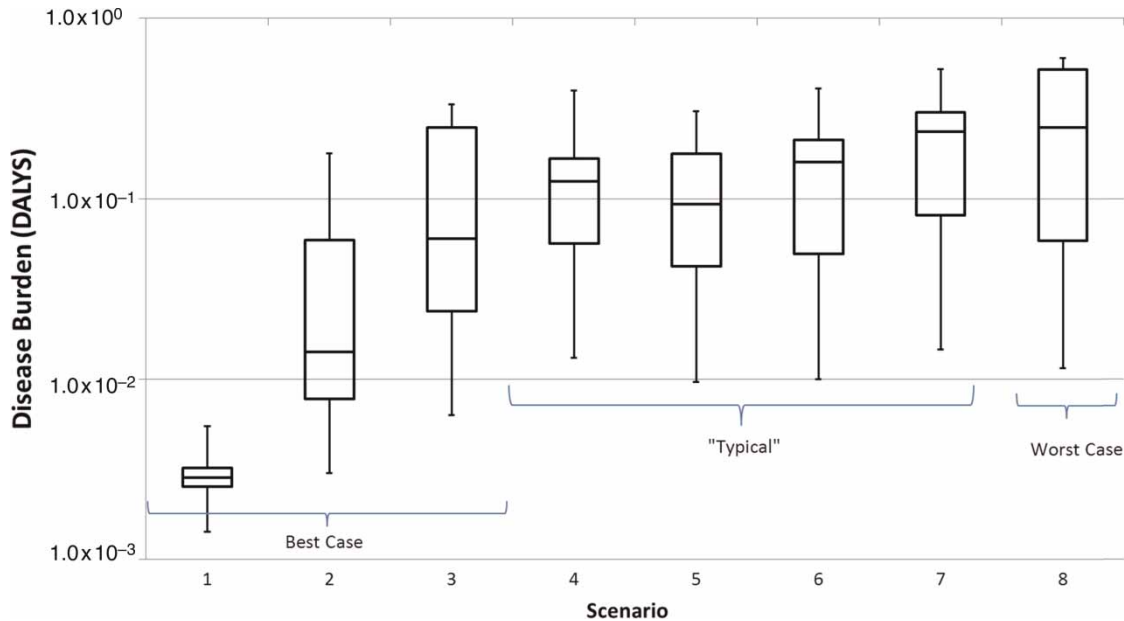


Figure 3 | Box and whisker plot representing the median, 25th and 75th percentile, maximum, and minimum estimates of risk for different water use scenarios.

The results of the QMRA analyses for the various scenarios are compared to piped water usage and the ‘best’, ‘worst’, and ‘typical’ water source combinations are identified.

The QMRA analysis revealed that the use of filtered water or boiled water stored in a kettle 100% of the year represented the ‘best’ case scenario, while 100% use of untreated shallow well water represented the ‘worst’ case scenario for residents in the resettled communities studied. Four ‘typical’ water use scenarios were identified. The results showed that these annual water usage combinations provided some risk reduction over exclusive use of surface water (‘worst case’); however, these ‘typical’ scenarios did not reach the lower levels of risk associated with the exclusive use of filtered water or boiled water (‘best case’) (Figure 3).

Estimation of disease burden by water source

Piped water

A 5-log removal rate for *E. coli* was applied to the influent water quality data to estimate the treated water quality in the piped distribution system. The average piped water risk level 2.9×10^{-3} is three orders-of-magnitude greater than the WHO recommended risk level. It should be noted that

the possible impact of interrupted service as pointed out by Hunter *et al.* (2009) was not addressed; however, due to recent improvements to Phnom Penh’s water treatment and distribution system (Chan 2009), this issue is not considered significant.

Untreated water supplies

Untreated rain, bottled, and tank water were found to have very similar levels of associated risk, even when examining the spread in the data. Since the use of these three water types carries similar levels of risk, households should prioritize increasing rainwater storage, which may be a more cost-effective option than bottled and tank water. However, it is not known how the rainwater quality may degrade with time. Degradation of rainwater quality is likely to correspond strongly to management practices; therefore, the risk associated with using untreated rainwater may increase over time (Schets *et al.* 2010). Untreated shallow well water has the highest level of risk of all the untreated water sources. The 75th quartile of untreated shallow well water exceeded the 75th quartile for all other water types. The mean risk associated with the consumption of shallow well water was 100 times greater than the risk associated with piped water consumption.

Boiled rain/tank water

The mean risk associated with the consumption of boiled rain/tank water stored in a 20 L bottle was the greatest for all water sources. A possible explanation for the increased risk for water stored in 20 L bottles is that these types of bottles are typically reused repeatedly by the households, and cleaning practices are likely to vary greatly between households. Contamination of boiled water when subsequently stored in 20 L bottles could potentially be a concern.

No treated water sources provided water of a mean level of risk within the same order-of-magnitude as piped water. Boiled water stored in a kettle, and boiled rain or tank water stored in a bucket/cooler, or bucket with a spigot were within one order-of-magnitude risk of piped water. It was expected that water drawn directly from kettles would have the lowest risk, as kettles are sterilized during boiling and the opportunity for contamination is decreased by not transferring the water to another vessel. However, it is not always practical to retain boiled water in kettles due to the low volume that may be stored. As a risk management strategy, it may be wise to prioritize the use of water stored in kettles and give it to higher risk individuals, such as young children, although some participants indicated that their young children do not like to drink warm water, and therefore will choose to drink cooler, untreated water, over recently boiled, warm water. In these cases, water stored in a bucket or cooler with spigot appears to be a good option, with an increase in mean risk of approximately 45% greater than that for water stored in kettles.

Boiled shallow well water

Boiled shallow well water stored in a bucket or cooler had a higher mean risk than boiled rain/tank water stored in a bucket/cooler with or without a spigot. However, boiled shallow well water stored in a bucket or cooler had a lower risk than rain/tank water stored in a small bottle or 20 L bottle. This result suggests that assuming effective boiling is being practiced, the resulting contamination, and risk associated with these water sources could be due to container maintenance/handling practices. Small plastic bottles and 20 L bottles tend to be reused frequently and are difficult to clean due to their small mouths.

Boiled shallow well water was found to have no statistically significant difference compared to boiled rain or tank water when stored in a kettle. This result also alludes to the fact that improper maintenance and water storage practices may be responsible for increased risk associated with water stored in a bucket or cooler instead of a kettle. It may be that the handling of untreated shallow well water, which has much higher levels of microbial contamination than untreated rainwater, may result in higher levels of contamination potential. For example, it is possible that some households may use the same storage containers for untreated and treated shallow well water. This finding may indicate that improper boiling and/or storage practices are being employed by households. The authors acknowledge that boiling is an effective method of treatment if done correctly; however, ineffective boiling and/or improper storage practices may increase risk.

Estimation of disease burden by water usage scenario

Countless possible drinking water usage combinations were used in the two communities; water usage changes over time and by season in many Cambodian communities. While rainwater is almost universally used as a primary water source in the rainy season, some households continue to use other sources such as shallow well water throughout the year. In the dry season, some Cambodian households have sufficient rainwater storage capacity and sufficient management practices to have rainwater supplies last throughout the dry season (Murphy *et al.* 2010). However, most peri-urban households switch to unimproved sources such as costly tank or bottled water or possibly heavily contaminated shallow well water. Shoulder periods or transition periods exist between rainy and dry seasons, when different water use scenarios will occur. These shoulder periods are not reflected in the present analysis. The rainy and dry seasons are each considered to be six months long.

Best (Scenarios 1–3) and worst case (Scenario 8) scenarios and a few ‘typical’ scenarios (Scenarios 4–7), based on evidence from the field were created and are described below. The QMRA analysis of the various scenarios shows a range of risk levels to be expected under different usage patterns. For example, while one household dispensed

drinking water from a kettle throughout the duration of the study, on average, households who used a kettle to dispense boiled rain or tank water did so approximately 40% of the time, with 60% of samples being drawn from a bucket or cooler. The ratios of boiled to unboiled samples and kettle to bucket-drawn samples were estimated from the water types collected at given households. The results of the risk characterizations for these scenarios are presented in Table 8.

Worst case scenario

Year round use of shallow well water is not common, given the widespread prevalence of rainwater harvesting for VS and PS; however, rainwater harvesting can be cost-prohibitive for some families. When comparing use of shallow well water year round (Scenario 8) with the seasonal use of untreated rainwater during half the year (Scenario 7 – typical scenario), the annual risk is decreased by nearly 25%. Consequently, the use of untreated shallow well water during the dry season has a large impact on the annual risk of illness.

Typical scenarios

Scenario 5 reflects a typical scenario of mixed storage use encountered in the field. With the use of boiled rain water during the rainy season (stored in a kettle 40% of the time; bucket 60% of the time), and boiled shallow well water in the dry season (stored in a kettle 20% of the time; bucket 80% of the time), the level of risk is nearly 2.5 times greater than could be expected if a kettle was used as storage at all times.

In the typical scenarios, boiling water shows some reduction of risk no matter what source water is involved. For example, when comparing Scenario 5 (all water is boiled, but boiled shallow well water is also used) with Scenario 4 (50% of water is left unboiled but only higher quality rain and tanker water are used), Scenario 5 gives a lower level of risk (11%) than Scenario 4.

Best case scenarios

The best case scenarios are based on 100% usage of piped water (Scenario 1), boiled water stored in a kettle (Scenario 2), or

filtered water (Scenario 3). While boiled water stored in a kettle may provide a reasonable alternative to piped water supplies in terms of risk level, few households achieve these levels of risk over the long term due to inconsistent practices. While one household in this study dispensed water using a kettle every sampling week, most only did so sporadically. Using filtered water 100% of the time did not provide a mean risk that was better than some of the ‘typical’ water usage scenarios, implying that the filters were not that effective or they were not being used and maintained properly. Murphy *et al.* (2010) found that improper cleaning practices associated with the use of CWPs increased levels of contamination in stored filtered water. Long-term use of CWPs and other filtration devices may differ based on how they are sold or otherwise distributed to communities. Achievement of either ‘best case scenario’ options (Scenarios 2 and 3) are unlikely to occur consistently in the long term. Consequently, the authors suggest that work be done to try to improve household awareness around appropriate maintenance and storage practices as well as try and minimize risk by evaluating the microbial risk of more ‘typical’ drinking water scenarios.

STUDY LIMITATIONS

When interpreting the results of this study, one must consider the study limitations. In this study, the authors were restricted to indicator organisms as the measure of microbial water quality; therefore, ratios of *E. coli* to pathogen(s), in this case ETEC, had to be assumed using data from the literature. There is definite uncertainty around this ratio; consequently, magnitudes of risk must be interpreted with caution. Nevertheless, for comparative purposes, since the same ratio was applied to all water types (including piped water), the magnitudes of risk difference between water sources and water scenarios would remain unchanged if this ratio were altered. These results are therefore still meaningful to compare risks between different water scenarios and consequently can be used to inform decision-making.

Another limitation is that given the case study nature of the present study, the sample sizes and collection period for each water type were relatively small. It is believed that the results are representative of the two communities in which

the samples were taken (internally valid). Lognormal probability distributions were assigned to the concentration inputs into the QMRA to represent the variability in water quality by water type. It is anticipated that accounting for this variability may represent the variability of water quality seen between peri-urban communities.

CONCLUSIONS

QMRA is a useful tool to compare risks associated with alternative water supply scenarios. In this paper, QMRA was used to examine the differing risks between 'best' and 'worst' case scenario water sources as well as 'typical' water source combinations used by peri-urban residents of Phnom Penh. Using QMRA in this way allows one to examine the risk due to existing water consumption practices and to compare the risk to possible intervention strategies. QMRA can be used to inform and better target interventions and be used as an advocacy tool with policy-makers to illustrate the increased health risk to households when resettled in a community without a piped water supply.

Upon moving, the majority of VS and PS residents experienced a drop in water quality and, therefore, a greater increase in predicted diarrheal disease risk. Boiling and storing water in a kettle was able to provide water of lower risk (only one order-of-magnitude higher than that of piped water), although at a higher cost and level of inconvenience. Most households do not maintain their boiled water supplies in ways to minimize risk; as a result, even households who do boil their water typically face risks of diarrheal disease from ETEC infection of one to two orders-of-magnitude higher than could be expected from piped water supplies.

Key findings from this study include

- Untreated rain, bottled, and tank water had similar levels of associated risk.
- Untreated shallow well water had the highest level of associated risk among untreated water sources.
- Boiled rain and tank water stored in a kettle, bucket/cooler, bucket with spigot were found to carry risk levels within one order-of-magnitude of the piped water available in Phnom Penh.

- Water stored in a 20 L bottle or small 500 mL bottle carried the highest level of risk, possibly due to poor cleaning practices between uses.
- Boiled shallow well water stored in a bucket or cooler had the largest spread of expected values of the boiled water samples, likely due to post-boiling handling practices.
- Boiled water stored in a kettle may provide water within one order-of-magnitude of risk of piped water supplies, but few households consistently use these methods.
- Using boiled rain water during the rainy season and boiled shallow well water in the dry season under a specific 'typical' storage scenario (mixed use of buckets and kettles for storage) results in a risk level nearly 2.5 times greater than could be expected if a kettle was used as storage at all times.
- The use of any untreated shallow well water greatly increases the annual risk level faced by a household.

The methodology presented to evaluate risk in water supplies and the relative risk expected when water sources and treatment methods are mixed over the course of the year are applicable to a broad range of scenarios.

More work is needed to improve access to high quality drinking water by the resettled residents of peri-urban Phnom Penh. There are opportunities to improve the present situation for resettled communities until piped water system access is extended to serve them. It is important that awareness is raised about problems with post-treatment and storage contamination of water. The use of untreated water from highly contaminated water sources can be expected to have a huge impact on overall risk levels and must be avoided. Consequently, interventions in the area of source water protection may help lower contamination levels of drinking water sources, such as hygienic handling and storage practices, the use of latrines, and containing livestock to defined areas away from water sources, and should be implemented.

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