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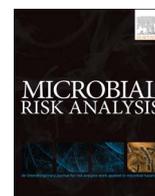
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Full length article

Probabilistic quantitative microbial risk assessment model of farmer exposure to *Cryptosporidium* spp. in irrigation water within Kumasi Metropolis-Ghana



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ABSTRACT

Cryptosporidium is a protozoan parasite which can be transmitted via food and water. Some studies have shown irrigation water to be routes of transmission for *Cryptosporidium* into the food chain, however, little information is known about *Cryptosporidium* levels in wastewater used for irrigation in the Kumasi Metropolis of Ghana. Kumasi and for that matter Ghana is not immune to the widespread practice of wastewater irrigation for farm produce in developing countries which has attracted attention of both, policy makers and academia. However, most previous studies of microbial risk assessment focus on the possible health effects and risk estimation for consumers of wastewater irrigated produce, whereas farmers who actually come into direct contact with the wastewater have received little attention. This study estimated the possible risk/diseases from farmer exposure to *Cryptosporidium*, a zoonotic pathogen causing gastroenteritis. The results indicate high positive levels of *Cryptosporidium* in the irrigation water, however, the levels of *Cryptosporidium* decreases during the rainfall seasons, risk assessment results show that, farmers face a higher risk of being infected by *Cryptosporidium* due to frequent exposure to wastewater. An adoption of a possible on-farm wastewater treatment option was found to reduce the risk of infection of the farmers. The results of this study highlight the need for a proactive policy to integrate a multi-barrier approach to reduce direct contact of farmers with wastewater for irrigation, to minimise risk of infection.

1. Introduction

Farmers cultivating lands in urban and peri-urban areas in most developing countries are known to use wastewater, mainly due to inaccessibility of fresh water. Wastewater is also a known public health concern, as a source of disease-causing microorganisms (Amoah et al., 2005; Drechsel et al., 2009; Keraita et al., 2002). Sources of water used by farmers for irrigation in urban and peri-urban areas include industrial, domestic, and agricultural wastewater. This may lead to contamination by oocysts of the human pathogen *Cryptosporidium* spp. that originates from infected humans and animals.

The protozoan parasite *Cryptosporidium* is a zoonotic pathogen capable of infecting the epithelial cell lining of the digestive tract of various host species including humans. The oocysts, which are environmentally robust, are responsible for several outbreaks of water-borne diseases worldwide, leading to serious implications for public health (Fayer et al., 2000; Mara and Nigel, 2003). Several studies on risk assessment with respect to consumption of vegetable produce grown on land irrigated with wastewater and the accidental ingestion of *Cryptosporidium*-infested wastewater have been reported (Mota et al., 2009; Teunis et al., 2002).

In Kumasi-Ghana, vegetable farming activities are mainly situated

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in low lands and are usually in close proximity to water bodies. It is estimated that, about 59 hectares of urban and peri-urban lands are invested into vegetable farming during the dry season, with a corresponding 48 hectares in the wet season (Keraita et al., 2014a). Studies (Drechsel and Keraita, 2014) have shown that, most farmers within the peri-urban centres rely on the use of wastewater for irrigation purposes. Moreover, other previous studies have shown that, there is high levels of *Cryptosporidium* spp. in these irrigated waters used by farmers in Kumasi (Petersen, 2015; Samposn, 2015) and again, several studies in Ghana (Adjei et al., 2003, 2004; Mor and Tzipori, 2008; Opintan et al., 2010; Eibach et al., 2015) have confirm human cases of *Cryptosporidium* spp. infections both in Kumasi and Accra.

In general, most Quantitative Microbial Risk Assessment (QMRA) measures of possible risk as a result of exposure to pathogens have focussed on health risks to consumers; however, less attention has been directed towards the risk to farmers exposed to wastewater used for irrigation in both, urban and peri-urban irrigation centres of food production. The aim of the study is to evaluate *Cryptosporidium* spp. concentrations in wastewater used by farmers in Kumasi, Ghana and the health risk associated with the accidental ingestion of wastewater by farmers who are frequently exposed.

2. Material and methods

This study was conducted on farms at four study sites, namely, Ahodwo, Chirepatre Estate, Twumduase, and Boadi (Fig. 1), all located within the Kumasi Metropolis of the Ashanti Region in Ghana. Water samples were collected between April 2014 and January 2015 and the permission to use these sites for the study was obtained from the Waste Management Department of the Kumasi Metropolitan Assembly as well

as the owners of the farms. The field study did not involve endangered or protected species nor was it conducted in any protected area.

2.1. Water sample collection and processing

All farms obtain irrigational water from different sources. Farm 1 in Ahodwo receives irrigational water from a stream-water using a pump where upstream wastewater from the Komfo Anokye Teaching Hospital (KATH) enters. Farm 2 in Chirepatre Estate receives irrigational water from two sources: a manually-dug well and a stream-water that is joined upstream, by effluents from a waste stabilization pond which also receives water from private houses in the vicinity, and run off from nearby green areas. Farm 3 in Twumduase receives irrigational water solely from 2 manually-dug wells. Farm 4 in Boadi receives irrigational water from a stream-water that is joined by various streams from surrounding communities (Fig. 1).

Collection of water samples was done twice per month from April 2014 to January 2015. Samples were taken within the two predominant weather seasons in Ghana, wet season (April–September) and dry season (October to March), samples were taken from all water sources per farms as described by Duhain (2011) and Chaidez et al. (2005). Volumes of 100l were filtered through polypropylene, 1-mm-poresize filters from each sampling point, samples were taken from the water source 20–30 cm beneath the water surface. Seventy-two (72) surface water samples were collected at each of the farms.

After sampling, each filter was placed in portable coolers for transport to the Biochemistry Department of Kwame Nkrumah University of Science and Technology (KNUST), Kumasi, for analyses using the U.S. Environmental Protection Agency Information Collection Rule Method (USEPA, 1995). For the purification of *Cryptosporidium*,

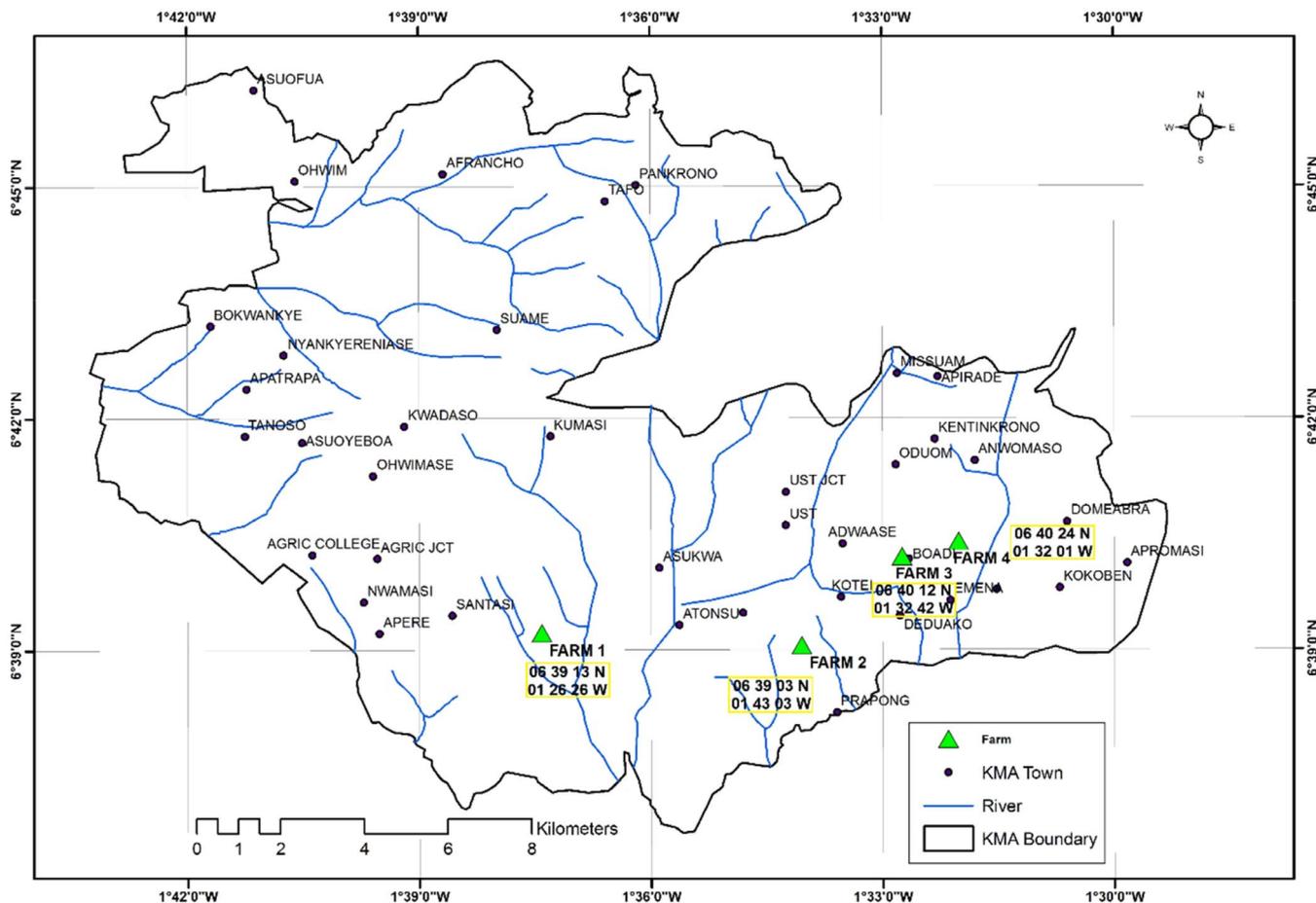


Fig. 1. Farm sites from where wastewater samples were collected in Kumasi, Ghana.

the filters were cut lengthwise and hand washed for 30 min with eluting solution as described in Mota et al. (2009). The eluting solution was concentrated by centrifugation, the supernatant was aspirated, and the filter sediment was resuspended in elution solution at the Department of Parasitology of the Noguchi Memorial Institute for Medical Research, University of Ghana. The concentrates were further purified by the flotation purification protocol. Finally, the sample was stained with a specific fluorescent antibody, and *Cryptosporidium* was identified based on size, shape, and fluorescence with an epifluorescent microscope and polymerase chain reaction (PCR). The presence of parasites was reported in numbers of (oo)cysts per 100 l of surface water sample. When the parasites were not detected, the parasite level was reported as less than the detection limit.

Paired sample test was used to investigate into the significance of *Cryptosporidium* spp. in the irrigation waters of the various farms for wet and dry seasons. Analysis of Variance (ANOVA) was also used to determine whether there were any statistically significant differences between the means of *Cryptosporidium* spp. concentration data among the farms, the data were assumed to be independent (unrelated from various farms and each season) groups. This procedure compares the means between the groups of interest and determines whether any of those means were statistically significantly different from each other. Specifically, it tests the null hypothesis: The *Cryptosporidium* spp. concentration means from the various farms were all statistically equal. The *Cryptosporidium* spp. concentrations from the various farms were then pooled after analysis of variance (ANOVA) showed no significant differences among the wastewater source from the farms as well as for the seasons.

2.2. Exposure assessment of farmers

Exposure assessment method addresses the likelihood of exposure to a hazard occurrence, which describes the quantity of hazard in the exposure (Ryu and Abbaszadegan, 2008). The exposure route was defined as the risk of exposure through accidental ingestion of wastewater as a result of irrigation processes by the farmers. The exposure pathway begins with hospital waste-water, domestic waste-water, Greenfield run-off, dug-well water entering a secondary water sources depending on the Farm practice where that particular farm gets its sources of water for irrigation. Concentration of other pathogens (viruses, bacteria, helminths, protozoans) have been found in these sources of water for irrigation in previous studies (Amoah et al., 2005, 2007; Silverman et al., 2013; Keraita et al., 2013; Drechsel et al., 2009). Upon entry into the water bodies, farmers fetch water manually with their watering cans from their water sources and applied on the vegetable plant (overhead spray irrigation) using the same watering cans. Predominantly, farmers use two watering cans to fetch water concurrently for irrigation, it is estimated that, one watering can as use in Ghana has a capacity of 15 l of water (Drechsel and Keraita, 2014), moreover, no protective clothing are worn during irrigation thereby exposing them to both direct accidental ingestion of wastewater, dermal contact as well as aerosol inhalation. A simple on-farm treatment options (three tank system, simple sedimentation and simple filtration) were used as a basic treatment for waters collected for irrigation to reduce the log reduction of pathogen concentration and offer some form of treatment to the waste-water as well as to reduce the risk of farmers (Keraita et al., 2014b; Amoah et al., 2011).

Exposure assessment was built with the use of stochastic (random) technique for the input parameters shown in Table 1. The pathogen dose (oocysts/day) ingested at each exposure for a farmer accidentally ingesting wastewater assumed to be contaminated was modified from earlier work by Mara et al. (2007) represented by,

$$d = C_{raw} \times R^{-1} \times I \times V \times 10^{-Q} \quad (1)$$

where d is the dose (oocysts/day) accidentally ingested by farmer, C_{raw} is the concentration of detectable (oo)cyst (oocyst/100 l) in wastewater;

Table 1
Input parameters for dose response model and risk characterisation.

Parameter	Values	Reference
No of <i>Cryptosporidium</i> oocysts/ 100 l of irrigation water (oocysts/l)	Range <52–105; geometric mean ^a 83.46	
Recovery efficiency (%)	Uniform ^b (15,20)	
Volume of irrigation water accidentally ingested/aerosol inhalation (ml/day)	Uniform (1,5)	Seidu et al. (2008), WHO (2006)
Percentage of infectious oocysts (%)	Point Estimate: 41	Ryu and Abbaszadegan (2008)
Three pond water treatment system (logs)	Uniform (1,2)	Amoah et al. (2011)
Simple sedimentation system (logs)	Uniform (0.5,1)	
Simple filtration system (logs)	Uniform (1,3)	

^a Geometric mean is defined as the n th root of the product of n numbers, thus with set of numbers x_1, x_2, \dots, x_n , the geometric mean is given as $(\prod_{k=1}^n x_k)^{1/n} = \sqrt[n]{x_1 \times x_2 \times \dots \times x_n}$.

^b Distribution: Uniform(minimum value, maximum value).

R is the recovery efficiency of the detection method used (%); I is the percentage of infectious oocysts (%); and V is the volume of wastewater accidentally ingested by farmers during irrigation(ml/day) (Mara et al., 2007; Petterson et al., 2007), and Q is the log reduction due to adopted on farm wastewater treatment option.

For this study, lower, upper and mean concentrations were used. The lower concentration was used to represent the detection limit of the method used to enumerate the *Cryptosporidium* spp. and the recovery efficiency (R) were determined from seeded limits conducted in the same laboratory and averaged 15% to 20% range of the *Cryptosporidium* spp.

Moreover, as previously reported (Mota et al., 2009; Ryu and Abbaszadegan, 2008; Hamilton et al., 2006), all oocysts detected were considered to be equally transferrable during accidental ingestion of water. Additionally, infectivity of the detectable oocysts was considered as 0.41 (Ryu and Abbaszadegan, 2008). Limited information is currently available regarding accidental ingestion of wastewater during irrigation, however, this was assumed to be uniformly distributed from 1 to 5 ml to account for the use of improvised equipment for irrigation practices (WHO, 2006) whilst the total exposure of farmers was also estimated to be a little over 2 months from sowing to harvest of vegetable produce. Thus, 60–70 days (Seidu et al., 2008) and quantified with uniform distribution. The total exposure estimated represented a single planting season, however, in this paper such an exposure is taken throughout the year, since farmers engaged in continuous farming due to the demand of vegetable products on the market. Q describes the log reduction for three scenarios of an adopted on-farm water treatment options which includes three tank system with log reduction of 1–2 logs, simple filtration with 1–3logs reduction and simple sedimentation 0.5–1logs of reduction (Amoah et al., 2011). All input parameters are indicated in Table 1.

2.3. Mathematical modelling approach

2.4. Dose response assessment

The dose response model used for *Cryptosporidium* infection was an exponential model given by (Furumoto and Ray, 1967)

$$P_{inf}(d) = 1 - \exp[-r*d] \quad (2)$$

where r is the dose parameter for *Cryptosporidium*, in this study, the value of r was taken to be 5.7×10^{-2} (Teunis et al., 2002). In Ghana human cases of *Cryptosporidium* spp. infections have been confirmed by several studies (Adjei et al., 2003, 2004; Opintan et al., 2010)

normally from diarrheic patients caused by *Cryptosporidium parvum* (Adjei et al., 2003; Mor and Tzipori, 2008; Eibach et al., 2015). Hence, the dose response model with its parameter was chosen to reflect on the prevalence of *Cryptosporidium parvum* in Ghana.

2.5. Risk characterisation

The annual probability of infection was estimated using the adjusted gold standard given as (Karavarsamis and Hamilton, 2010)

$$P = 1 - \prod_{i=1}^N (1 - P_{inf,i})^n \tag{3}$$

where $P_{inf,i}$ is the i th weekly probability of infection caused by *Cryptosporidium* and N is the number of periodic infection probabilities in a year defined with a uniform distribution of 40–52 weeks and n represents the period over which the assumption of constant daily infection probability is extended which is taken as 7 days. and P is the annual risk of infection.

To account for variability and uncertainty in the parameters, different parts of the model were subjected to Monte-Carlo simulation of 100,000 iterations with hypercube sampling for the annual probability of infection and a sensitivity analysis were done. All the models were constructed in Microsoft Excel using the @ Risk 7.5 (Palisade Corporation) software add-on to Excel.

3. Results and discussion

3.1. Results from *Cryptosporidium*

3.1.1. Prevalence of *Cryptosporidium oocyst* in sample irrigation water

An overall prevalence of 66.67% (48/72) for *Cryptosporidium oocyst* positive presence was observed among the irrigation water samples. *Cryptosporidium oocysts* were detected in all the water samples used in the various farms as shown in Table 2. A prevalence of 55.6% (10/18), 61.11% (11/18), 77.78% (14/18) and 72.22% (13/18) oocyst positives were observed in water samples for Farm 1, Farm 2, Farm 3 and Farm 4 respectively There was no significant difference in detectable oocyst during the two seasons ($p = 0.525$). as well as among the farms (ANOVA), though it was established that, while all farms had positive water samples in all seasons. Wet season which comes with rainfall seemingly lowered the concentration of oocysts in water at all the farms hence the lower mean *Cryptosporidium spp.* data as compared to the dry season though no significant observable differences ($p > 0.05$) were recorded.

The probable risk from various microbial concentration levels was assessed and the estimate of probability of infection per event/day as well as the annual probability of infection were estimated. Four scenarios were adopted which includes the irrigation practices using

Table 2
Prevalence, average and test of significant difference.

	Number tested		Number positive (%)	
Farm 1	18		10(55.56)	
Farm 2	18		11(61.11)	
Farm 3	18		14(77.78)	
Farm 4	18		13(72.22)	
Total	72		48(66.67)	

	Wet season	Dry season	p-Value	Pooled data
Farm 1	55.57 ± 5.09	76.86 ± 21.62	0.06	68.33 ± 19.47
Farm 2	63.29 ± 15.52	82.71 ± 19.88	0.08	72.84 ± 19.66
Farm 3	69.14 ± 23.07	88.85 ± 17.84	0.21	78.66 ± 21.14
Farm 4	61.43 ± 21.68	78.43 ± 20.03	0.22	67.58 ± 21.03

For detail analysis refer to supplementary sheet.

Table 3
Risk assessment for farmers with disaggregated data of various farms.

Risk scenarios	Farm 1 Median	Farm 2 Median	Farm 3 Median	Farm 4 Median
Raw wastewater	5.44×10^{-4}	5.89×10^{-4}	6.18×10^{-4}	5.21×10^{-4}
Three tank system	1.59×10^{-5}	1.73×10^{-5}	1.81×10^{-5}	1.52×10^{-5}
Simple sedimentation	9.24×10^{-5}	1.01×10^{-4}	1.05×10^{-4}	8.84×10^{-5}
Simple filtration	5.04×10^{-6}	5.45×10^{-6}	5.69×10^{-6}	4.82×10^{-6}

untreated wastewater and three on-farm treatment option (simple filtration, simple sedimentation and three tank pond system), the upper limit of detection was used as the worst case scenario for all scenarios in the pooled data analysis.

3.1.2. Risk assessment with disaggregated data from farms

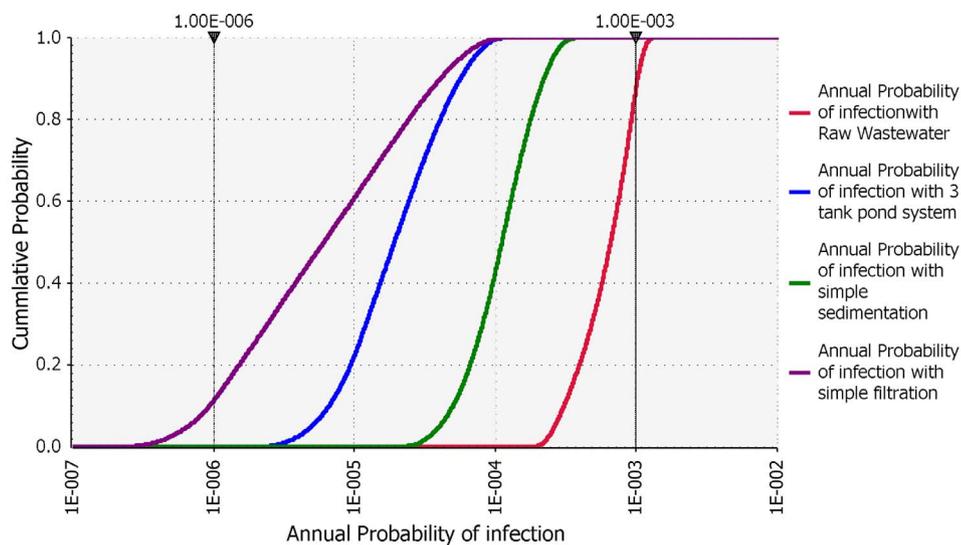
The annual median risk of farmers for each of the farms were found to be 5.44×10^{-4} , 1.59×10^{-5} , 9.24×10^{-5} and 5.04×10^{-6} for raw wastewater, three tank system, simple sedimentation and simple filtration respectively for Farm 1 (Table 3). All the other farms also recorded higher risk (greater 1 out of a million per year) for farmers, it is observed that, farm 3 poses a higher risk, followed by farm 2 and farm1 with farm 4 having the least possible risk.

3.1.3. Risk assessment with aggregated (pooled) data of all farms

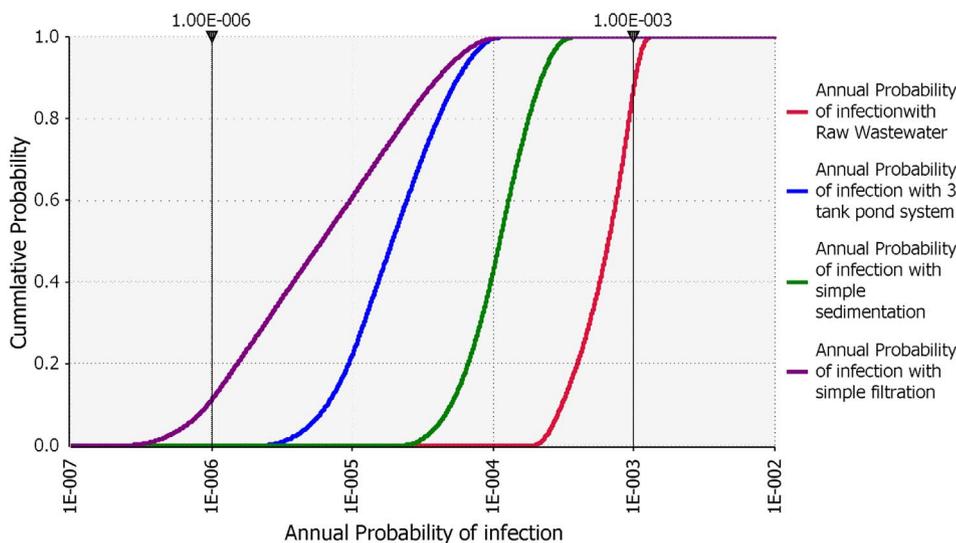
The daily median (50th percentile, Fig. 2) risk of infection were found to be 6.34×10^{-6} , 1.85×10^{-7} , 1.07×10^{-6} and 5.82×10^{-8} and the mean infection were also found to be 6.38×10^{-6} , 2.49×10^{-7} , 1.19×10^{-6} , and 1.37×10^{-7} for Raw Wastewater, Three tank system, Simple Sedimentation and Simple Filtration respectively for the lower concentration, the daily median risk of infection for the upper concentration were 1.28×10^{-5} , 3.73×10^{-7} , 2.16×10^{-6} and 1.18×10^{-7} and the mean infection were 1.29×10^{-5} , 5.04×10^{-7} , 2.42×10^{-6} , and 2.77×10^{-7} for raw wastewater, three tank system, simple sedimentation and simple filtration respectively, whereas the geometric median concentration level also recorded a daily risk probable estimation of 1.01×10^{-5} , 2.96×10^{-7} , 1.72×10^{-6} and 9.38×10^{-8} and the mean infection were 1.02×10^{-5} , 4.01×10^{-7} , 1.92×10^{-6} , and 2.20×10^{-7} for raw wastewater, three tank system, simple sedimentation and simple filtration respectively (Table 4, Fig. 2). As expected, the mean and median risk estimates across all scenarios for the lower concentration were lower than the upper concentration risk estimates and that of the geometric mean concentration for all the scenarios falls within the upper concentration risk estimates and the lower concentration risk estimates.

The mean and the 50th percentile daily risk estimates for the upper concentration were found to be lower than WHO benchmark of 1.0×10^{-6} when the three tank system and the simple filtration on-farm treatment methods are adopted, nevertheless, the simple sedimentation falls short of less than 1 log of reduction whereas the estimates of the raw wastewater did not meet the daily risk estimate benchmark.

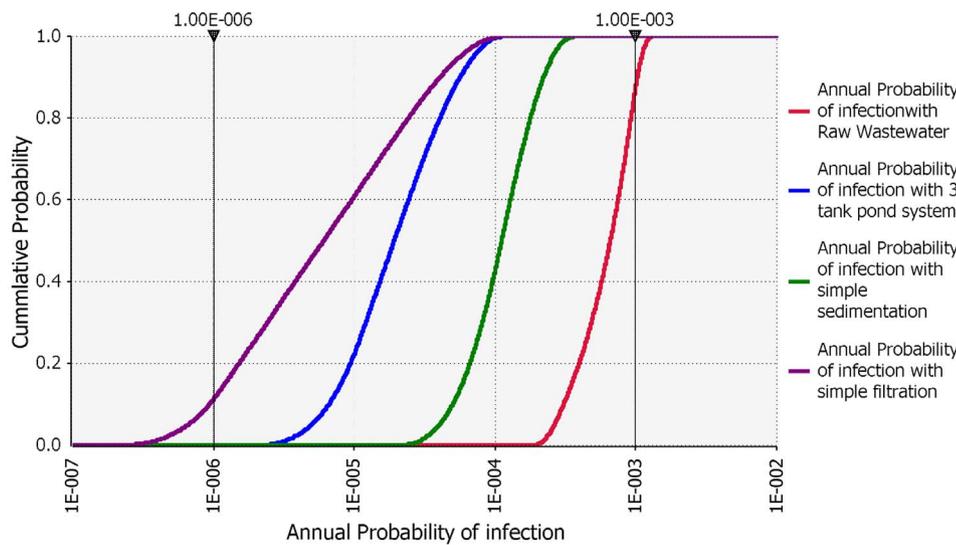
The estimated annual median risk of infection for lowest detectable *Cryptosporidium oocysts* concentration ranges from 3.78×10^{-6} to 4.09×10^{-4} and the mean ranges from 8.90×10^{-6} to 4.15×10^{-4} for all scenarios, the upper concentration limit has median risk estimate ranges from 7.63×10^{-6} to 8.2×10^{-4} and its mean values range from 8.37×10^{-4} to 3.27×10^{-5} as well as the geometric mean oocysts concentration with annual median risk ranges from 6.08×10^{-6} to 6.59×10^{-4} whereas its mean values ranges from 6.65×10^{-4} to 2.60×10^{-5} (Table 5). The mean and the 50th percentile probable risk estimate of the upper concentration were all higher than the WHO benchmark of 1.0×10^{-6} irrespective of the on-farm treatment option adopted. These findings did not show any significant deviation from the



(a) lower pathogen concentration risk estimates



(b) mean pathogen concentration risk estimates



(c) upper pathogen concentration risk estimates

Fig. 2. Annual cumulative risk assessment, (a) *Cryptosporidium* spp. lower concentration (b) *Cryptosporidium* spp. lower concentration (c) mean *Cryptosporidium* spp. concentration.

Table 4
Probability of infection per exposure for farmer.

Risk Scenarios	Range							
	52 oocysts/100l				105 oocysts/100l			
	Mean	5th Percentile	50th Percentile	95th Percentile	Mean	5th Percentile	50th Percentile	95th Percentile
Raw wastewater	6.38×10^{-6}	2.54×10^{-6}	6.34×10^{-6}	1.05×10^{-5}	1.29×10^{-5}	5.12×10^{-6}	1.28×10^{-5}	2.11×10^{-5}
Three tank system	2.49×10^{-7}	4.83×10^{-8}	1.85×10^{-7}	6.69×10^{-7}	5.04×10^{-7}	9.75×10^{-8}	3.73×10^{-7}	1.36×10^{-6}
Simple sedimentation	1.19×10^{-6}	3.88×10^{-7}	1.07×10^{-6}	2.45×10^{-6}	2.42×10^{-6}	7.85×10^{-7}	2.16×10^{-6}	4.95×10^{-6}
Simple filtration	1.37×10^{-7}	6.42×10^{-9}	5.82×10^{-8}	5.40×10^{-7}	2.77×10^{-7}	1.30×10^{-8}	1.18×10^{-7}	1.10×10^{-6}

Annual probability risk scenarios	Geometric mean: 83.46 oocysts/100l			
	Mean	5th Percentile	50th Percentile	95th Percentile
Raw wastewater	1.02×10^{-5}	4.07×10^{-6}	1.01×10^{-5}	1.68×10^{-5}
Three tank system	4.01×10^{-7}	7.74×10^{-8}	2.96×10^{-7}	1.08×10^{-6}
Simple sedimentation	1.92×10^{-6}	6.22×10^{-7}	1.72×10^{-6}	3.95×10^{-6}
Simple filtration	2.20×10^{-7}	1.01×10^{-8}	9.38×10^{-8}	8.73×10^{-7}

disaggregated data of the various farms.

Wastewater irrigation as a practice for substituting freshwater for irrigation purposes might be a good alternative, if wastewater treatment measures are put in place to ensure achieving an acceptable pathogen level for both unrestricted (Unrestricted irrigation is defined as permitting irrigation of all crops) and restricted (Restricted irrigation is defined as permitting irrigation restricted to salad crops and vegetables that are eaten raw) irrigation as described in the WHO policy document (WHO, 2011). Not surprisingly, farmers in developing countries engaging in non-mechanised farming have direct contact with the wastewater as a result of the use of improvised equipment for irrigation, it is therefore predictable that, the estimate of median annual probability of infection for upper detection level was higher than the recommended benchmark of infection of 1.0×10^{-6} by Signor and Ashbolt (2009) or the WHO standard of 1.0×10^{-6} (Mara and Sleight 2009; Signor and Ashbolt, 2009).

The WHO guideline states that, 'if the overall burden of diseases from other exposures is very high, setting a less stringent level of acceptable risk of 1×10^{-4} or 1×10^{-5} threshold may be more realistic' as was argued by Mara and Hamilton (2010). In dealing with risk estimates for farmers who use improvised equipment and are much higher of being directly exposed to pathogen infested wastewater, it is important to stick to the more stringent benchmark of 1×10^{-6} , nevertheless, if one is to go by the argument made by Mara and Hamilton (2010) for a less stringent health target of acceptable risk of

1×10^{-5} (Mara and Sleight, 2009; Mara and Hamilton, 2010; Mara et al., 2010), then the probable median risk estimate value to the health target were not met in all scenarios in this study. Should be noted that, with the adoption of other on-farm practices such as wearing protective gear during irrigation, a proper irrigation method combine with the on-farm wastewater treatment options could reduce the annual risk of infection to an acceptable level, nevertheless, the farmers in the study do not practice such other practices (wearing protective gear, using proper irrigation methods such as drip irrigation).

3.2. Sensitivity analysis

Sensitivity analysis was used to identify the model parameters with significant impact on the risk output. It was observed that the annual probability of infection was very sensitive to *Cryptosporidium* spp. concentration in irrigational water, the on-farm water treatment method, daily accidental ingestion of wastewater and the total exposure (frequency exposure) to wastewater for each irrigation period (Table 6). These factors recorded a positive direct relationship with the risk estimate for the farmers and identify input parameters that can influence in mitigating the risk that farmers are exposed to, with regard to wastewater used for irrigation. The sensitivity analysis indicated that, key parameter for the risk estimate was the initial level of *Cryptosporidium* spp. contamination level in wastewater and had a strong positive relationship with the risk estimate for all scenarios.

Table 5
Yearly risk of *Cryptosporidium* infection of farmers associated with accidental ingestion of wastewater for irrigation in Kumasi-Ghana.

Risk scenarios	Range							
	52 oocysts/100l				105 oocysts/100l			
	Mean	5th Percentile	50th Percentile	95th Percentile	Mean	5th Percentile	50th Percentile	95th Percentile
Raw wastewater	4.15×10^{-4}	1.64×10^{-4}	4.09×10^{-4}	6.86×10^{-4}	8.37×10^{-4}	3.33×10^{-4}	8.26×10^{-4}	1.38×10^{-3}
Three tank system	1.62×10^{-5}	3.12×10^{-6}	1.19×10^{-5}	4.37×10^{-5}	3.27×10^{-5}	6.33×10^{-6}	2.42×10^{-5}	8.83×10^{-5}
Simple sedimentation	7.79×10^{-5}	2.51×10^{-5}	6.94×10^{-5}	1.60×10^{-4}	1.57×10^{-4}	5.07×10^{-5}	1.40×10^{-4}	3.23×10^{-4}
Simple filtration	8.90×10^{-6}	4.16×10^{-7}	3.78×10^{-6}	3.51×10^{-5}	1.80×10^{-5}	8.37×10^{-7}	7.63×10^{-6}	7.12×10^{-5}

Annual probability risk scenarios	Geometric mean: 83.46 oocysts/100l			
	Mean	5th Percentile	50th Percentile	95th Percentile
Raw wastewater	6.65×10^{-4}	2.64×10^{-4}	6.59×10^{-4}	1.09×10^{-3}
Three tank system	2.60×10^{-5}	5.02×10^{-6}	1.92×10^{-5}	7.03×10^{-5}
Simple sedimentation	1.25×10^{-4}	4.03×10^{-5}	1.11×10^{-4}	2.57×10^{-4}
Simple filtration	1.43×10^{-5}	6.58×10^{-7}	6.08×10^{-6}	5.68×10^{-5}

Table 6
Sensitivity analysis (Spearman's correlation coefficient).

Parameter	Spearman's correlation coefficient			
	Raw wastewater	Three tank pond system	Simple sedimentation	Simple filtration
<i>Cryptosporidium</i> spp. concentration	0.54	0.52	0.61	0.59
Volume of irrigation water accidentally ingested	0.97	0.51	0.77	0.29
Recovery rate	0.19	0.10	0.14	0.05
Total exposure	0.11	0.05	0.07	0.03
Treatment with three tank system		−0.84		
Treatment with simple sedimentation			−0.60	
Treatment with simple filtration				−0.95

3.3. Assumptions and uncertainty associated with the model

Quantifying the sources of uncertainty as well as variability is essential for QMRA. In this study, although *Cryptosporidium* oocyst concentration data from the sampling sites do not represent a comprehensive survey of wastewater for irrigation by farmers within Ghana, nevertheless, it gives a fair perspective of *Cryptosporidium* contamination in wastewater.

Recovery efficiencies reported during the experimental work were not uniform across all experimental procedures, hence recovery efficiency estimation theory by Petterson et al. (2007) was applied to take into consideration uncertainty surrounding the different efficiencies. In addition, the QMRA model did not include *Cryptosporidium* oocyst inactivation owing to the assumption of direct accidental ingestion of wastewater, leaving no interval for direct contact of oocysts with the environment or sunshine, to initiate or continue the process of inactivation. It is known that oocyst inactivation is mostly influenced by sunshine (Reinoso and Bécarea, 2008). Furthermore, the dose estimation is a considerable source of uncertainty in this study, and did not account for resistance due to temporary immunity of farmers as a result of continuous exposure to the wastewater. Possibly, such acquired immunity of farmers is likely to reduce the risk of infection; however, studies on acquired immunity of farmers to *Cryptosporidium* infection are not currently available, studies have indicated higher levels of risk of gastroenteritis for households which irrigate their farm with wastewater (Cifuentes, 1998), nevertheless, there are reports of limited cases of gastroenteritis infection risk due to acquired temporary immunity (Linnemann et al., 1984). There is a lack of comprehensive study on the actual amount of *Cryptosporidium* spp. that could be ingested through daily accidental ingestion of wastewater, which is due to improvised equipment used in developing countries; this represents a source of uncertainty that could lead to underestimation of risk to farmers that could have been 1 or 2 logs of magnitude higher.

In this study, the QMRA level of annual risk of infection of farmers did not meet the WHO benchmark; hence, reduction of the risk by a higher oocyst concentration reduction in wastewater is required.

3.4. Risk management strategies and recommendations

The risk from wastewater irrigation depends on several factors such as irrigation method, wastewater treatment options, and requirement of a multi-barrier approach, as outlined by WHO (2006). Given the widespread practice of wastewater irrigation in Ghana, there is the need for better wastewater regulation that will protect farmers and reduce their contact with *Cryptosporidium* oocysts. This approach may need a more proactive management approach to help minimise the risk due to exposures. The WHO guidelines for wastewater reuse provide a detailed structure for building country-specific reuse guidelines that include various multi-barrier approaches that could be flexible and consistent with local policy, beliefs, and culture. The multi-barrier approach could be focused in areas such as reducing *Cryptosporidium* spp. and daily accidental ingestion of wastewater by farmers by incorporating

appropriate measures to minimise the direct contacts with wastewater as these tend to have a positive correlation. Farmers are important stakeholders in the agricultural industry and potential on-farm management options together with irrigation methods and appropriate farm equipment for irrigation purposes can assist in mitigating the risk of *Cryptosporidium* spp. exposure during irrigation of farm products.

This study recommends some risk management strategies that could be implemented to reduce potential exposure to Farmers during irrigation. WHO's multiple barrier approach supports a range of further options for the management of risks from pathogens on farm such as:

- A minimal (low-cost) wastewater treatment option (1–2 units pathogen reduction).
- Drip irrigation (2–4 log units pathogen reduction).

Other measures can include the following:

- Protecting the adopted on farm treatment option from external sources such as birds and other animals which can re-contaminate the treated water.
- Using the appropriate water-can for irrigation such as capped water-can raised less than 0.5 m above the ground to reduce splashing and hence reduce exposure to aerosol accidental ingestion as described by Amoah et al. (2011).
- Permitting sunlight to reach the treatment water option to assist in photo-inactivation of potentially harmful pathogens.

Therefore, it is essential to prioritise Hazard Analysis Critical Control Point (HACCP) initiatives to reduce the risk level that farmers are exposed to while using wastewater for irrigation.

4. Conclusion

QMRA is a powerful tool for risk assessment of farmers directly exposed to wastewater during irrigation. We estimated the annual probable risk of infection of farmers with lower limit mean concentration, upper limit mean concentration, and the geometric mean concentration of the pathogen concentration of *Cryptosporidium* oocyst data from four (4) different vegetable farms which use wastewater for irrigation; Four (4) different scenarios were presented. The results show a higher risk of infection in all scenarios and did not meet the threshold of 1×10^{-6} benchmark. Risk of infection were higher for estimates with upper limit concentrations, followed by geometric mean oocyst and then lower limit concentration. Due to this, a multi-barrier approach with a local policy guideline is a necessity to help minimize the associated risk of infection of farmers using wastewater for irrigation.

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Supplementary materials

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