



# Influence of sewage treatment plant effluent discharge into multipurpose river on its water quality: A quantitative health risk assessment of *Cryptosporidium* and *Giardia*<sup>☆</sup>



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## ABSTRACT

Sewage treatment plants (STPs) are one of the sources of pathogens discharged into surface water. An investigation was carried out over the duration of 12 months in Henan Province, China, to evaluate the health influence of municipal wastewater effluent discharge on water quality of the receiving water. A discharge-based quantitative microbial risk assessment (QMRA) was employed, taking into account the vegetables consumption habits of the Chinese, population subgroups with different immune statuses and ages, to evaluate the incremental disease burden from agricultural irrigation and swimming exposure scenarios associated with increased concentration of the protozoan *Cryptosporidium* and/or *Giardia* in the receiving river. The results shown that all the STP influent samples contained *Cryptosporidium* and *Giardia* with average density of 142.31 oocysts/L and 1187.06 cysts/L, respectively. The QMRA results demonstrated that the estimated additional health burdens due to discharged effluent for both parasites were slightly violated the threshold of  $10^{-6}$  DALYs per person per year set by WHO. Mitigation measures should be planned and executed by season since more disease burdens were borne during hot season than other seasons. The sensitivity analysis highlighted the great importance of stability of STP treatment process. This study provides useful information to improve the safety of surface water and deduce the disease burden of the protozoa in Henan Province and other region inside and outside China.

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## 1. Introduction

Sewage discharge is one of the problems in both developed and developing countries. It contributes to a compounded unstable aquatic ecosystem, including but not limited to oxygen demand and nutrient loading of the receiving watershed, promoting toxic, and algal blooms (Gonzalo and Camargo, 2013; Li et al., 2016; Auberttheau et al., 2017; Hilario Garcia et al., 2017). In addition, sewage effluent may be of public health significance, especially if effluent is discharged into water that is subsequently used for

drinking, recreation, or agricultural purpose (Robertson et al., 2006; Zafar et al., 2017). Effluents from industrial sources, generally because of their chemical pollution, are associated with non-communicable disease (Zafar et al., 2017), while those from municipal wastewater would contribute to infectious disease (King et al., 2017; Mok et al., 2014). The infectious agents associated with municipal wastewater are those found in the domestic sanitary waste of the population.

Although a variety of pathogens have been identified in raw sewage, relatively few types of pathogens seem to be responsible for most of the waterborne diseases caused by sewage-original pathogens (Seto et al., 2016; Mead et al., 1999). Among them, protozoan parasites such as *Cryptosporidium* and *Giardia* are the key types of pathogens due to their high infectivity and high resistance to water treatment and disinfection (WHO, 2009). Consequently any additional sources of *Cryptosporidium* oocyst and *Giardia* cyst in receiving water are highly undesirable.

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In fact, *Cryptosporidium* and *Giardia* were responsible for the most prevalent worldwide waterborne infection producing diarrhea. In immunodeficiency person such as human immunodeficiency virus (HIV)-infected patients, this infection is more common and can be life-threatening (Hunter and Nichols, 2002). During the time period from 2004 to 2010, at least 60.5% (120) and 35.1% (70) of worldwide waterborne outbreaks of human diseases caused by *Cryptosporidium* and *Giardia*, respectively, and most of the outbreaks occurred in developed countries (Baldursson and Karanis, 2011). This may be a distorted picture of the global conditions which was attributed to the improved reporting from regions of high economic and sanitation status (WHO and UNICEF, 2017). The infection prevalence and outbreak incidence in developing countries probably most affected by both of the parasites are not able to identify or provide any reports of outbreak events due to the lack of surveillance systems (Efstratiou et al., 2017a). In Thailand, which belongs to developing countries like China, the reported data of diarrheal diseases incidence were suspected to be under-reported and the causal agents of diarrheal diseases were undiscovered (Yotthanooi and Choonpradub, 2010). It has been estimated that nearly 2% of adults and 6%–8% of children in developed countries worldwide, while nearly 33% of the people in the developing countries have had giardiasis ([www.cdc.gov](http://www.cdc.gov)).

Many factors including the multiple exposure routes reflecting living and sanitation conditions, and contamination of the aquatic ecosystem by the high environmental burden, have been contributed to the high infection caused by *Cryptosporidium* and *Giardia* (Squire and Ryan, 2017). Between January 2007 and December 2008, 96.7% of waterborne cryptosporidiosis outbreaks in the United States of America were associated with recreational water other than drinking water (Brunkard et al., 2011). On the other hand, the health risks of *Cryptosporidium* and *Giardia* infection associated with agricultural irrigation have become one of major concerns of the world, particularly in developing countries (Mok and Hamilton, 2014; Aiello et al., 2013).

Quantitative microbial risk assessment (QMRA) has been widely employed to predict the human health risks associated with exposure to infectious pathogens in drinking water (Xiao et al., 2012a), surface water (An et al., 2011; Adell et al., 2016), and reclaimed water (Zhang et al., 2015; Ryu et al., 2007). Given the concentrations of pathogen in receiving water are often 'non-detect' with limits of detection, the lack of appropriate input data for pathogen concentrations at the point of exposure limits the application of QMRA. To meet this challenge, McBride et al. (2013) developed a discharge-based QMRA that uses pathogens detected in discharges into flowing inland waters rather than relying on measurements in receiving waters themselves to assess the risk of seven pathogens in recreational waters. Later, the discharge-based QMRA was employed in drinking water management (Sokolova et al., 2015).

Effluents of sewage treatment plant (STP) have been increasingly used for irrigation, recreational impoundments, and wetland reconstruction in many arid regions of the world (Ma et al., 2016). In China, there are about 1.33 million hectares of land irrigated directly and indirectly with wastewater (Jiménez, 2006) and 10.5–21.6% of people swim in river (An et al., 2011). Considering that China is the world's most populated country, with 74,000 HIV-positive persons (UNAIDS, 2011), there may be millions of people exposed to health risks from water. As far as we know, to date there have not been any studies on the microbial risks attendant with effluent discharging into river in China. The aim of this research was to investigate the health influence of municipal wastewater effluent discharges into river on its water quality. For this purpose, the concentrations of *Cryptosporidium* oocysts and *Giardia* cysts in influent of STP were surveyed for a year, and a discharge-based

QMRA combined with hydrologic modeling was employed, taking into account the vegetables consumption habits of the Chinese, population subgroups with different immune statuses and ages, to evaluate the incremental disease burden from agricultural irrigation and recreational exposure associated with increased concentration of these protozoa in river. The uncertainties in these attributable risk estimates are discussed as well as their implications. The methodology and results of this study will be useful in better evaluating and reducing the burden of protozoan infection associated with wastewater in Henan Province and other region inside and outside China.

## 2. Material and methods

### 2.1. Study site description

The STP is located in the county of Wuzhi, Henan Province, China (Fig. 1). The treatment system at the STP consists of a preliminary treatment (rotating screens and aerated grit chamber), oxidation ditch, and disinfection with chlorine. The population served by this STP is approximately 150,000 inhabitants. This plant treats around 24,000 m<sup>3</sup> of domestic sewage per day, which is discharged into a small receiving water body, and finally about 10% of the effluent reaches the nearby *Qinhe* River.

The *Qinhe* River is the main source of agricultural irrigation in the local farmland, and is recreational water for some people in the hot season. In 2015, the water flow of the river varied between 3.14 and 10.82 m<sup>3</sup> per second, with an average of 6.98 m<sup>3</sup> per second. The lowest and highest water level of the *Qinhe* River usually occurred in the dry season (November–March) and rainy season (June–August), respectively.

### 2.2. Hazard identification

Bacteria, viruses, and protozoans are the major groups of sewage pathogens. Most of these pathogenic microorganisms could be controlled by the application of disinfection with chlorine. However, *Giardia* cyst and *Cryptosporidium* oocyst are moderately even highly resistant to both environmental stress and chemical disinfection (WHO, 2009). Furthermore, these protozoa cannot be completely removed by the conventional wastewater treatment. Consequently, *Cryptosporidium* and *Giardia* in the discharged effluent were considered to be the pathogens most likely to pose a threat, and were selected as indicators, for this study, to assess the public health influence of effluent on river water quality.

### 2.3. Exposure assessment

#### 2.3.1. Concentration of parasitic pathogens

Compared to the much more removed and dilute concentration expected in effluent and receiving water, pathogens were generally present at higher concentration in the influent, increasing the likelihood of their detection without the need for large-volume sampling (McBride et al., 2013). The concentrations of *Cryptosporidium* oocyst and *Giardia* cyst in sewage influent of the STP were monitored every two weeks from April 2015 to March 2016 (n = 24). Each of monitoring days, 50 mL of sample was collected and concentrated by centrifugation at 1500 × g for 10 min. Then, oocysts and/or cysts in the pellet were separated from debris by flotation on Percoll-sucrose gradients, labeled with fluorescein isothiocyanate conjugated anti-*Cryptosporidium* and anti-*Giardia* monoclonal antibodies (Waterborne, Inc. New Orleans, LA) as well as 4'6 diamidino-2-phenyl indole (DAPI), and were examined microscopically, as previously described by Xiao et al. (2012a). This analytical method based on microscopic examination can detect as

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