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DETECTION OF GIARDIA CYST AND TAENIA EGGS IN RIVER WATER SAMPLES COLLECTED FROM ALETO RIVER IN ELEME LOCAL GOVERNMENT AREA, RIVERS STATE, NIGERIA

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ABSTRACT

This study aimed at detecting Giardia cyst and Taenia eggsin river water samples collected from Aleto River in Eleme Local Government Area, River State, Nigeria, to check for potability and purity of water.Water samples of river water were collected from two points (downstream and midstream) of Aleto river. Labelled clean sample bottles with tightly-fitted lids were used to collect water samples from the river and stored in coolers with ice packs to maintain their temperature and transportation to the laboratory. Collected water samples were transferred into sterile vial tubes and centrifuged at 2000 rpm for five minutes, after which the supernatant fluids were discarded, while the sediments smeared using spatula on labelled slides stained with specific dyes, using the wet mounts, trichome stain technique, and modified Ziehl–Neelsen techniques, and viewed under the light microscope to identify the presence of parasites.This study showed the presence of ten trophozoites of Giardia lamblia and ten eggs of Taenia species isolated from the river water samples collected. These findings highlight the potential health risks associated with waterborne parasites and emphasizes the importance of effective water quality management to safeguard public health. Furthermore, the observation of parasites underscores the need for comprehensive water quality assessment and monitoring. This study highlights the contamination of the river with parasitic organisms and urgently needs effective water management strategies to mitigate the risks to public health. Vital interventions encompass enhancing sanitation practices, curbing fecal contamination, and augmenting water quality surveillance mechanisms.

Keywords: *Giardia* species, microscopy techniques, parasites, public health, river water, *Taenia* species

INTRODUCTION

Giardiasis is a worldwide distributed parasitic infection which has been included in the "Neglected Diseases Initiative" since 2004, they are heterogeneous group of parasites that occur mostly in developing countries, which are related to poverty, and lack of access to public and medical services (Rosado-García *et al*., 2017). Efstratiou*et al.*, 2017 reported one hundred and forty-two waterborne diarrhea outbreaks caused by *Giardia* species

from 2011 to 2016, from a limited number of developed countries. In most middle- and low-income countries.There are limited reports of *Giardia* outbreaks due to the lack of documentation of water parasitic protozoa in water supply by the government (Efstratiou*et al.,* 2017). Humans are exposed to *Giardia* infection by the ingestion of cysts present in contaminated water or food (Adam *et al*., 2016). *Giardia* cysts are highly resistant in the aquatic environments and to some disinfectants, like chlorine concentration applied in water treatment plants for water purification (Carmena, 2010). However, it is relevant to determine the presence of these cysts in water sources intended for human consumption, to reduce the risk of infection.

Taenia are tapeworm species of medical importance in humans that may lead to health and economic burden (Jansen, 2018). Humans are the main definitive hosts of these three zoonotic *Taenia* species, namely; *T. saginata, T. solium and T. asiatica* (WHO, 2020). Infections with *T. solium*and *T. asiatica* are considered to be among the neglected tropical diseases, and especially for infections caused by the former, which requires adequate measures for control and elimination because these parasites can cause cysticercosis in humans. Infection with a tapeworm (taeniosis) are usually asymptomatic (Laranjo-González *et al.,* 2016) with some exceptions (Bekraki and Hanna 2016). *Taeniasolium*, *Taeniasaginata* and *Taeniaasiatica*, are also known as neglected foodborne parasites which are of public health and economic importance because of their disease transmission (Alvarez-Rojas *et al*., 2018). *Taenia* tapeworms excrete thousands of eggs which contaminates the environment or water bodies through unhealthy sanitary practices, such as open defecation, thus exposing humans and animals to possible risk of infection after consuming infested water (Jansen *et al*., 2021). In developing countries, inadequate sewage treatment is generally considered as a huge contribution to cattle infections by *T. saginata*, as animals become infected by ingesting the eggs from contaminated pastures after flooding or from access to surface water (Laranjo-González *et al.,* 2016). However, in low-income countries, humans contaminate the environment (soil, crops and water) with *Taenia* eggs present in faeces due to poor hygienic standards and the lack of latrines (Arriola *et al.,* 2014). In general, contamination of food, soil and water can increase the risk of human infection with *T. solium* and other intermediate hosts of *Taenia* (Dalu*et al*., 2011: Federer *et al*., 2016).

Intestinal parasitic infections are regarded as neglected infectious diseases because they are of public health concerns, which cause illnesses such as abdominal disorders, retarded growth, impairment of cognitive skill in young children, and also serious economic problems in most subtropical and tropical countries (Becker *et al*., 2013). These diseases affect millions of people, especially those in endemic areas, ranging from 30-60% of the population (Ammoura*et al.,* 2010).

Drinking water or potable water also known as improved drinking water is said to be any water that is free from physical, chemical, biological or radiological form of contamination (Gyang *et al*., 2017). Absent, inadequate, or inappropriately managed water and sanitation services expose individuals to preventable health risk (Pam *et al.,* 2018). Biological contaminants such as bacteria, viruses, fungi, protozoa and helminthes constitute the major cause of food-borne and water-borne diseases with varying degrees of severity ranging from mild indisposition to chronic or life-threatening illness or both (Ani and Itiba, 2015). Parasitic contaminations which lead to infections have detrimental impact on host nutritional status in several ways, they can depress appetite and food intake, compete for micronutrients, or cause blood loss, resulting in the loss of iron, diarrhea, vomiting, dehydration, weight loss and growth retardation (Omolade and Zanaib, 2017).

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In Africa, children under the age 5 suffer from infectious diarrhea which leads to death (Walker *et al*., 2012). Unsafe water usage and lack of adequate as well as substantial level of sanitation and hygiene increase the transmission of pathogenic organisms which causes diarrhea related diseases (WHO,2015). Two major enteric protozoan parasites, *Cryptosporidium* and *Giardia* are associated with diarrheal diseases (Feng *et al*., 2011), with *Cryptosporidium* being the most common worldwide (WHO,2015). Many rural homes in Africa and over three hundred million people in Sub-Sahara Africa have limited access to potable water and use untreated water for domestic purpose which includes bathing, drinking, swimming, cooking and washing, which expose them to waterborne pathogens (WHO, 2015). The lack of adequate water and lack of awareness / educational programmes, predisposes many African communities to the two major water related protozoan diseases, Cryptosporidiosis and Giardiasis(Sente *et al*., 2016). Squire and Ryan (2017) researched on *Cryptosporidium* and *Giardia* in Africa and opined that poverty was a major limiting factor to accessing safe drinking water, and most individuals in Africa have limited access to privately owned water resources. Many researchers have reported the presence of *Cryptosporidium* oocysts and *Giardia* cysts in African water sources including irrigation water in Burkina Faso (Kpoda *et al*., 2015), stream, well, spring and lake in Cameroon (Ajeagah, 2013), packaged drinking water in Ghana and South Ethiopia (Damitie*et al*., 2020), tap water, drinking water treatment plants, tanks in Egypt (El-Kowrany *et al*., 2016).

Waterborne diseases accounts for 80% of illnesses in developing world, killing a child every 8 seconds. It is also believed that hospital beds are occupied by people suffering from water borne diseases due to polluted drinking water (Isirimah, 2003).This study aimed at detecting *Giardia* cyst and *Taenia* eggsin river water samples collected from Aleto Riverin Eleme Local Government Area, Rivers State, Nigeria, to check for potability and purity of water.

MATERIAL AND METHODS

Description of the Study Area

The study area comprises of a freshwater river surrounded by forest vegetation in Aleto communityin Eleme Local Government Area (LGA) in Rivers State, Nigeria. River water samples were collected between the boundary of Rivers state and Abia state, between latitude 4.8070 and 4.8092 N and longitude 7.1012 and 7.1249 E. The river flows beneath the Elemebridge, and it is contaminated with oil spillage due to pipeline vandalization and breakage. The indigenes of this community carry out domestic, sanitary and anthropogenic activities by the river side, which includes having their baths, open defecation, washing of clothes and dishes, as well as fishing activities.

Study Design

Two liters of river water was collected from the sampling station in duplicates (downstream and midstream) each month for a period of six months (March, 2022 to September, 2022) from Aleto river in welllabelled sterile bottles and transported to the laboratory, the Scan Doctor Medical Diagnostic Center, Port Harcourt, Rivers State, for microscopy and parasite identification. Water samples were transferred into labelled 10 ml of sterile vial tubes and centrifuged at 2000 rpm for five minutes. After which the supernatant fluid was discarded, the sediments were smeared with a spatula on labelled slides and stained with dyes, which were viewed under the microscope for examination, to identify the presence of parasites in the water samples, following standard procedures and protocols to ensure accurate results (Kwakye-Nuako*et al.*,2007).

Sample Collection and Analysis

Water Sample Collection

i. **Sampling Stations**: River water samples of were collected from Aleto river from

two points, the downstream and the midstream due to the activities that take place in the river, such as washing, bathing, open defecation and fishing, which were potential pollution sources.

ii. **Sampling Equipment**: The sampling bottles used to store the water samples from the river and equipment for sampling were washed and rinsed to prevent contamination of samples. All sample bottles where properly labelled to indicate the day, time and location of sampling.

iii. **Sampling Techniques:** Before collecting the samples, gloves and safety precaution such as personal protective equipment (PPEs) were worn. Clean sample bottles with a tight -fitting lid was used to collect water samples from the river. The bottle was submerged below the water surface to a depth of 0.3m below the river surface and allowed to fill completely before capping it underwater to minimize air bubbles (Environmental protection (water) policy, 2009)

iv. **Transportation of samples to the laboratory:** The water samples were stored in coolers with ice packs to maintain their temperature during transit and handled with care during transportation to the laboratory, to prevent spills or contamination.

v. **Storage**: Upon arrival to the laboratory, Scan Doctor Medical Diagnostic Center, Port Harcourt, Rivers State, stored water samples were refrigerated and kept at specific temperatures to preserve their quality until analysis.

vi. **Analysis:** Microscopic analyses were performed on the water samples to identify the presence of parasites, following standard procedures and protocols to ensure reliable results.

Parasitological/Microscopic Analysis:

In the laboratory, the collected river water samples were transferred into 10 ml of sterile vial tubes and centrifuged at 2000 rpm for five minutes (Kwakye-Nuako*et al.*,2007). After which the supernatant fluid was discarded, while the sediments smeared with a spatula on labelled slides and stained with specific dyes, and viewed under the light microscope for examination, to identify the presence of parasites in the water samples, using the following methods below:

Method 1:Wet Mounts: A drop of saline solution was placed on a smear sediment placed on a labelled slide, and mixed with a drop of iodine solution using an applicator stick, and covered with a cover slip, then examined for parasites under a microscope, using X10 and X40 objective lenses (WHO, 1991).

Method 2: Trichome Staining Technique: A drop of each sediment was deposited onto a labeled slide and spread into a thin, uniform smear using an applicator stick. These slides were then fixed in Schaudin's solution for thirty minutes. Subsequently, the slides were immersed once in iodine-alcohol solution for one minute, then twice in 70% ethanol for one minute each. Following this, they were stained with tetrachrome stain for eight minutes and de-stained with acetic acid alcohol solution for five seconds. The slides were dipped twice in 95% ethanol for two seconds each and one dip in absolute ethanol for one minute before being transferred into xylene for two minutes. Finally, the slides were observed under a light microscope using a high-power objective lens of X 100 (Kaplan, 1992: Cheesbrough, 2005a).

Method 3: Modified Ziehl–Neelsen Technique: Thin and uniform smears of the sediment was prepared on labelled slides, using the trichome stain technique, and then fixed in methanol for two minutes. These slides were subsequently stained with cold carbolfuchsin for five minutes and differentiated in sulfuric acid for another five minutes. Following a five-minute rinse under running tap water, the slides were counterstained with 0.25% malachite green. After another five-minute rinse under running tap water and drainage to dry, the slides were examined using a high-power objective lens

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of X100, with oil immersion for better visibility of parasites (Shimizu, 1992: Cheesbrough, 2005b).

RESULTS

A total of twenty – four liters (24,000 ml)of river water samples were collected from Aletoriver, 2 liters of river water from each sampling points, downstream and midstream of the river on monthly intervals, for six months as shown in Table 1. After thorough microscopic examination to identify the presence of parasites in the water samples, two parasites (protozoan and helminthes) were identified. These parasites included: ten trophozoites of*Giardia lamblia* and ten eggs of *Taenia* species as shown in figure 1.

Parasite Detection and Prevalence Rate

Table 1: The prevalence rate of parasites in the river water samples, collected every month.

Parasite	Sampling points	1st Month	2nd Month	3rd Month	4th Month	5th Month	6th Month	Total number of parasites
Giardia	Downstream	3	4	2	1	-	-	10
lamblia	Midstream	-	-	-	-	-	-	-
Taenia	Downstream	4	4	2	-	-	-	10
species	Midstream	-	-	-	-	-	-	-

Images of Parasite Isolates

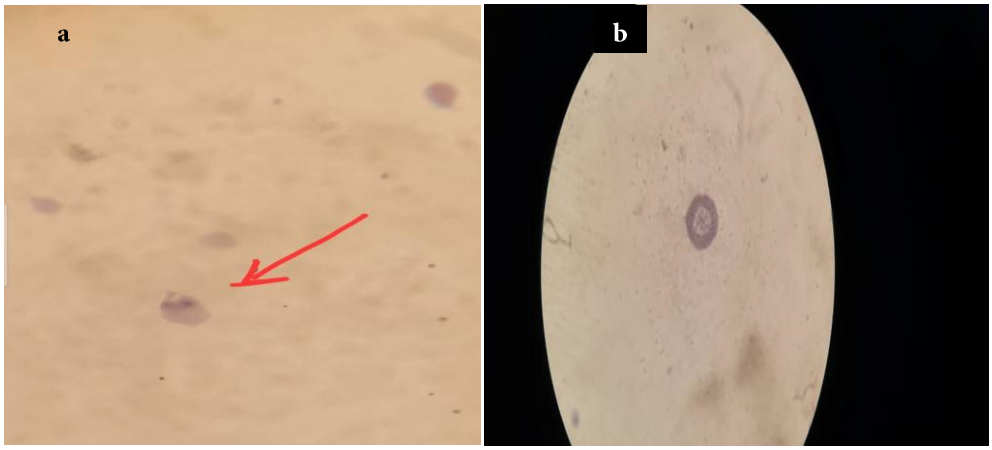

Fig. 1: Some organisms found in the water sample. **a)** Trophozoite stage of Giardia species **b)** Egg of Taenia species using trichomeand modified Zielhl – Neelsenstaining techniquesunder a light microscope

DISCUSSION

The results of this study revealed the presence of two parasites, namely *Giardia lamblia* and *Taenia* species, in water samples collected from Aletoriver. These findings highlight the potential health risks associated with waterborne parasites and underscore the importance of effective water quality management to safeguard public health.

The presence of *Giardia lamblia* in the water samples is consistent with previous studies conducted in different regions of Nigeria (Gyang*et al.*, 2017; Odikamnoro*et al.*, 2016; Oyibo*et al.*, 2016). *Giardia* infection, typically transmitted through ingestion of contaminated water or food, poses a significant threat to human health (Adam *et al.*, 2016). Similarly, the detection of *Taenia* species eggs in the water samples is alarming, as it indicates fecal contamination and raises concerns about the potential transmission of tapeworm infections (Jansen *et al.*, 2021).

The findings of this study corroborate earlier research, such as the study conducted by Gyang*etal.* (2017), which identified

protozoan parasites, including *Giardia lamblia*, in water sources contaminated by human and animal fecal matter. Additionally, Odikamnoro*et al.* (2016) reported the presence of various waterborne parasites, including *Giardia lamblia*, in water sources affected by improper sanitation practices. Similarly, Oyibo*et al.* (2016) found *Giardia* species and *Cryptosporidium* species in water samples collected from different sources, highlighting the role of unsanitary practices and animal contamination in waterborne parasite transmission.

The high prevalence of parasites in pond water, as observed in the study by Abdullahi *et al.* (2018), underscores the need for improved water sanitation measures, particularly in areas where open water bodies are susceptible to contamination. The presence of *Giardia lamblia* and *Entamoeba histolytica* cysts in the water samples further emphasizes the importance of addressing fecal contamination and promoting safe water practices to prevent waterborne diseases.

Furthermore, the observation of colored particles and debris in the river water samples underscores the need for comprehensive water quality assessment and monitoring. Poor sanitary practices, erosion, and influx of surface water into water sources contribute to water pollution and compromise water safety for human consumption.

CONCLUSION

In conclusion, this study highlights the widespread contamination of water sources with parasitic organisms and underscore the urgent need for effective water management strategies to mitigate the risks to public health. Vital interventions encompass enhancing sanitation practices, curbing fecal contamination, and augmenting water quality surveillance mechanisms. These proactive measures are imperative in safeguarding access to clean and potable water for communities inhabiting affected regions. Moreover, concerted endeavors from local authorities are indispensable to fortify preventive strategies aimed at mitigating intestinal parasite infections. This necessitates comprehensive health education initiatives targeting rural populations to improve community hygiene standards and foster overall well-being among residents.

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