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OCCURRENCE OF PROTOZOAN PARASITES IN WASTEWATER USED FOR IRRIGATION IN ILORIN

Badir-deen, M. S., Ojo, S., Lateefat, B. M., Iysa, O. M. and Kehinde, A. O.
Department of Zoology, Faculty of pure
and Applied Sciences, Kwara State University Malete.

Abstract

Parasitic infections pose a significant global health threat, particularly in developing countries where access to clean water and proper sanitation is often limited. Municipal wastewater, which is frequently reused for irrigation, often contains infective stages of parasites, posing a substantial risk to human health. Despite this, there is a lack of data on the protozoan parasites in wastewater in Ilorin Metropolis, Kwara State. This study addresses this knowledge gap on the protozoan parasites in wastewater used for irrigation. Water samples were collected from various irrigation sites and examined microscopically for protozoan parasites. The results revealed that 20.28% of the infective stages of protozoan parasites, mainly *Giardia, Cyptosporidium and Entamoeba, were* recorded per litre of water. However, using clean water from borehole will reduce the level of prevalence as lower prevalence was significantly observed at Zango which served as the control site for the study. A knowledge, attitudes, and practices (KAP) assessment further revealed that all respondents used contaminated water and were exposed to open defecation. While most farmers were aware of waterborne parasites and treated water before use, fewer buyers had such awareness, though many claimed to wash the products before resale. These findings indicate a substantial public health risk, particularly for communities relying on these water sources for agricultural irrigation. The study underscores the urgent need for enhanced wastewater treatment processes, sensitisation and better management practices to mitigate the health risks associated with wastewater reuse in agriculture. Continuous monitoring, awareness programme and the development of more effective treatment technologies are recommended to ensure the safety of agricultural practices in regions reliant on wastewater irrigation.

Keywords: Protozoan parasites, Wastewater, Irrigation

1.0 Introduction.

The sustainable management of water resources is a pressing global issue, particularly in regions experiencing rapid urbanisation, population growth, and climatic variability. In response to increasing water scarcity, the use of untreated or partially treated wastewater for irrigation has become a widespread practice in many low- and middle-income countries, including Nigeria (Jiménez & Asano, 2008; Qadir *et al*., 2010). This practice is most common in urban and peri-urban areas where access to clean water is limited, and agricultural land is often located near urban wastewater discharge points. While the reuse of wastewater offers socioeconomic benefits such as increased food production, cost savings, and nutrient recycling, it also poses several health and environmental risks if not properly managed (Drechsel *et al*., 2010). As reported by Chau *et al.* (2014), wastewater can contain harmful pathogens, including bacteria, viruses, and protozoan parasites commonly *Cryptosporidium*, *Isospora and Cyclospora*, which can pose significant health risks to farmers and consumers.

Intestinal protozoan parasites are significant contributors to the global burden of diarrheal diseases and are transmitted primarily through the faecal-oral route, often via contaminated water, food, or hands (Fayer *et al*., 2000). Protozoan parasites like *Giardia* and *Cryptosporidium* pose a significant public health risk because their resistant infective stages can persist in the environment, cause severe illness even at low doses, and contaminate crops irrigated with wastewater, especially affecting vulnerable populations (Robertson *et al*., 2010; Efstratiou *et al*., 2017).

Detecting protozoan parasites in wastewater is difficult due to their small size and the need for

Corresponding Author: **Badir-deen M. S.**
Department of Zoology,
Faculty of pure and Applied Sciences,
Kwara State University Malete.
badirdeen.salaudeen@kwasu.edu.ng

specialized techniques, and conventional treatment processes often fail to eliminate their resilient cysts and oocysts. In Ilorin, there is limited data on the presence of these parasites in irrigation water, as most studies focus on bacterial indicators. This is concerning, because protozoa can survive treatment and remain on crops, posing serious health risks, especially in the absence of adequate awareness. Addressing this gap is essential for protecting public health and shaping effective wastewater reuse policies. This study aims to assess the prevalence of protozoan parasites in irrigation wastewater in Ilorin Metropolis, evaluate the associated risks, and promote safer agricultural practices aligned with WHO guidelines and the One Health approach.

MATERIALS AND METHODS

Study Area

The study was conducted at selected sites in Ilorin, Kwara State. The state is located between latitude 8°05´N and 10°15´N and longitude 2° 73´E and 6°13´E in the North Central, Nigeria (Fig 1). Ilorin is the capital city of Kwara state with an area of 36,825km (Nigerian Investment Promotion Commission (NIPC), 2024).

Kwara State is within the forest-savanna region of Nigeria with a tropical climate characterized with a prolonged dry season (October to March) and rainy season (April to September) and with an average annual rainfall of 1200mm and mean temperature of 26.2 ? . The population is about 3,390,330 (NIPC, 2024). Youths and middle age make up the larger part of the population and open defecation is a common attitude. The selected sites are Harmony, Kuntu, Odota and Zango.

Sample and Data Collection

Wastewater samples were collected three times per week and at different intervals of the day [7-10 am (Morning), 1-3 pm (Afternoon) and 5-7 pm (Evening)] from selected water bodies using 2 litres sterile bottles. The samples were preserved in 5% Formaldehyde. All preserved samples were taken to the Department of Zoology laboratory, Kwara State University, Malete for analysis

Personal observation of water was done discreetly, and 50 copies of pre-tested structured questionnaires aimed at assessing knowledge, attitude and practices of farmers and buyers of the produce were administered.

Laboratory Analysis

The Bailenger technique was employed for the analysis of water samples. Approximately 1 liter of each water sample was filtered to remove debris and then allowed to sediment for 12 to 24 hours. After sedimentation, the supernatant was carefully decanted, and the remaining sediment was mixed with aceto-acetic buffer solution adjusted to pH 4.5. A few drops of the mixture were placed on a clean microscope slide, covered with a coverslip, and examined under a light microscope at 4× and 10× magnifications. Protozoan parasites were identified based on their morphological features using standard parasitological keys.

Data Analysis

The results obtained were analysed using the Microsoft Excel package 2013 and SPSS Statistical package version 29.0.2.0. The data were compared using the ANOVA test with a level of $p \leq 0.05$ at 95% Confidence Interval.

RESULTS

Fig 2 shows the average cyst concentration of protozoan class in wastewater at the study sites. The average concentration of protozoan class cysts in the wastewater was 20.28 cysts/oocysts per litre. The identified protozoan cysts belonged to the classes Apicomplexa, Mastigophora and Sarcodina, with a higher prevalence of Mastigophora. The chart shows that the cysts of Mastigophora and Sarcodina were higher at Kuntu than any other sites while Harmony recorded a higher concentration of Apicomplexans. Zango, the control site, recorded a lower concentration for all the 3 protozoan classes.

Protozoan prevalence across different periods of the day shows the highest mean concentration of infective stages at the early hour between 7.00 am and 10.00 am for majority of the parasites per millilitre of water; *Giardia* (3.08), followed by *Entamoeba* (2.54) and *Cryptosporidium* (2.31). In contrast, *Trichomonas* and *Cyclospora* had lower mean concentrations of 1.54 and 1.33 respectively. The standard deviations suggest that *Entamoeba* had a higher variability (1.71) between 7.00 am and

10.00 pm as compared to other protozoan types. The protozoan concentrations across different times of the day were statistically significant between 1.00 pm and 4.00 pm ($p = 0.012$) and 5.00 pm and 7.00 pm ($p = 0.004$), (Table 1).

Table 2 shows the prevalence of the protozoan parasites at different times of the day in all the sampling sites. Cyst concentrations at different sampling sites across various times of the day varied significantly ($p = 0.032$) with the prevalence of protozoan parasites in the wastewater used for irrigation. At Odota, the mean cyst concentration in the early hour of the day was 2.17 Cysts/ml but decreased slightly in the afternoon and evening. At Harmony, cyst concentration was consistent throughout the day, with a mean of 1.83 to 1.96 Cysts/ml while at Kuntu, mean concentrations, particularly in the morning was 2.38 Cysts/ml. The lowest mean concentration of 1.75 Cysts/ml was recorded at Zango.

The knowledge assessment, attitude and practices of farmers and buyers is shown in table 3. All the respondents admitted being the user of dirty water for irrigation and, are exposed to open defecation. More than 50% of the farmers responded to be aware of the parasites in water and treating water before use. On the other hand, buyers' awareness of parasites in water was less than 50% while more than 50% responded to washing the produce before reselling them.

DISCUSSION

A recent study carried out in Ilorin, Kwara State, Nigeria, uncovered clear differences in the levels of protozoan contamination across various irrigation sites and times of day. On average, about 20.28 protozoan cysts/oocysts were found in every litre of wastewater sampled. Among the different types of protozoa observed, flagellated forms (Mastigophora) were especially dominant at the Kuntu site, while Apicomplexa were more common at the Harmony site. Interestingly, the Zango site which used borehole water instead of wastewater had significantly fewer cysts across all protozoan types. This clearly shows how the source of irrigation water can greatly influence the level of contamination, a trend that has been reported in several parts of the world (Efstratiou *et al*., 2017; Al-Gamal *et al*., 2019).

These findings are not unique to Ilorin. Similar studies from other countries have reported high levels of protozoa in irrigation water, especially when wastewater is used. For example, in Mexico, researchers found *Giardia* and *Cryptosporidium* cysts ranging from 17 to over 1,600 cysts per 100 litres in both irrigation and vegetable-washing water (Chaidez *et al*., 2005). In the United States and Central America, a multi-site study reported *Giardia* in about 60% of irrigation samples, with an average of 559 cysts per 100 litres, while *Cryptosporidium* appeared in 36% of the samples (Thurston-Enriquez *et al*., 2002). These numbers closely resemble what was found in Ilorin, confirming that untreated wastewater poses a global risk for spreading protozoan parasites.

In Ilorin, *Giardia* was the most frequently detected protozoan, especially in samples collected in the early hour of the day. Although time-of-day differences in protozoan presence haven't been deeply studied in Nigeria, similar patterns have been seen elsewhere. For example, researchers in temperate countries have reported that *Giardia* levels in surface water often rise in late summer, likely due to runoff and increased human activity (Robertson *et al*., 2006). This suggests that the timing of sample collection can influence the presence of protozoan parasites, and that these fluctuations may be due to both environmental and human factors.

The Zango site, which used borehole water, had much lower protozoan counts, an encouraging finding that supports previous research in Nigeria. For instance, a study in Akure (Ondo State) showed that borehole water was free of protozoan parasites, while surface and well water sources were often contaminated with *Giardia*, *Entamoeba*, and *Cryptosporidium* (Simon-Oke *et al*., 2020). However, even at Zango, some infective stages were still detected, possibly due to contamination from soil or human and animal waste running off into water storage or distribution systems.

In addition to testing water, the study also looked at how much farmers and buyers knew about protozoan contamination and how they responded to the risks. Unfortunately, many of them, especially the buyers, had little awareness of the dangers. This isn't surprising. Even globally, studies have shown that knowledge alone doesn't always lead to better practices. In Pakistan, for example, farmers who used wastewater for irrigation were often aware of the risks but continued using it due to lack of access to clean water or better alternatives (Ensink *et al*., 2006). This highlights the need for not just education, but also real support like infrastructure and policy that can help communities make safer

choices.

Other countries have reported similar experiences. In Nepal, Shrestha *et al*. (2016) found high levels of protozoa in wastewater used for vegetable farming. In Turkey, Sağlam *et al*. (2022) showed that untreated wastewater had significantly more protozoan contamination than treated water. Comparable studies in Egypt (Hemati *et al*., 2022) and China (Huang *et al*., 2023) have also confirmed that even after treatment, protozoan parasites can remain a major concern in agricultural water.

Perhaps most worrying is that these parasites don't just stay in the water, they make their way into our food. In one study from Nigeria, Agbalaka *et al*. (2019) found *Giardia* and other parasites on vegetables irrigated with contaminated water. *Giardia*, which is passed in faeces and spreads through soil or water, causes *Giardia*sis—a disease that leads to chronic or severe diarrhea.

CONCLUSION

This study successfully investigated the prevalence and types of protozoan parasites in wastewater used for irrigation in Ilorin metropolis, Kwara State. The findings revealed significant variations in the types and prevalence of protozoan parasites across different sites, with high prevalence of *Giardia*, *Entamoeba* and *Cryptosporidium* species in wastewater samples. The research also highlighted the critical role of water sources in determining the concentration of protozoan cysts, with borehole water exhibiting lower levels of contamination compared to wastewater.

Moreover, the study assessed the knowledge, attitudes, and practices (KAP) of farmers and buyers regarding the use of contaminated water for irrigation. The results indicated a gap in awareness among both groups, with a notable lack of understanding of the health risks associated with protozoan contamination. This underscores the need for targeted educational interventions to improve the handling and use of irrigation water, thereby reducing public health risks.

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Figures



Fig 1:Map showing Sampling Sites

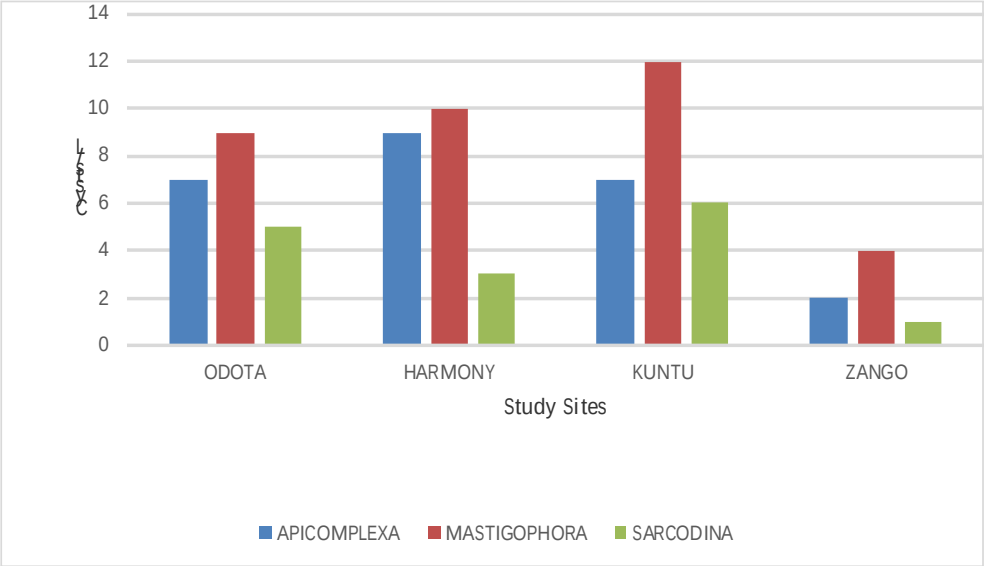


Fig 2: Average egg concentration of protozoan in waste waters at study sites.

Tables

Table 1: **PROTOZOAN PREVALENCE AT DIFFERENT TIME OF THE DAY**

Protozoan Types		Cysts/ml (Morning)	Cysts/ml (Afternoon)	Cysts/ml (Evening)
Cryptosporidium	Mean	2.31	1.62	2.08
	Number	13	13	13
	Std. Deviation	0.86	0.96	1.04
Giardia	Mean	3.08	2.54	2.62
	Number	13	13	13
	Std. Deviation	0.86	0.88	1.26
Entamoeba	Mean	2.54	2.15	2.15
	Number	13	13	13
	Std. Deviation	1.71	1.82	1.52
Trichomonas	Mean	1.54	1.31	1.46
	Number	13	13	13
	Std. Deviation	0.88	0.75	0.88
Cyclospora	Mean	1.33	1.42	1.83
	Number	12	12	12
	Std. Deviation	0.78	1.00	0.72
Isospora	Mean	1.75	1.33	1.42
	Number	12	12	12
	Std. Deviation	0.75	0.89	0.79
Total	Mean	2.11	1.74	1.93
	Number	76	76	76
	Std. Deviation	1.17	1.17	1.12
	P-value	0.399	0.012	0.004

Table 2: AVERAGE CYST CONCENTRATION AT DIFFERENT SAMPLING SITES

Sampling site		Cysts/ml (Morning)	Cysts/ml (Afternoon)	Cysts/ml (Evening)
Odota	Mean	2.17	1.67	2.04
	Number	24	24	24
	Std. Deviation	1.31	1.13	1.27
Harmony	Mean	1.83	1.79	1.96
	Number	24	24	24
	Std. Deviation	1.17	0.78	0.75
Kuntu	Mean	2.38	2.04	2.13
	Number	24	24	24
	Std. Deviation	1.06	1.40	1.12
Zango	Mean	1.75		
	Number	4		
	Std. Deviation	0.96		
Total	Mean	2.11	1.74	1.93
	Number	76	76	76
	Std. Deviation	1.17	1.17	1.12

Table 3: **ASSESSING KNOWLEDGE, ATTITUDE AND PRACTICES OF FARMERS AND BUYERS OF THE PRODUCE.**

ITEMS		
KINDS OF CROPS	Vegetable. 31 (62%)	Fruits. 19 (38%)
KINDS OF IRRIGATING WATER	Dirty 50 (100%)	Clean water 0 (0%)
OPEN DEFECATION IN T HE WATER	Yes 50 (100%)	No 0 (0%)
DO FARMERS TREAT THE WATER BEFORE USE	Yes 40 (80%)	No 10 (20%)
FARMER'S AWARENESS O F PARASITES IN WATER	Aware 30(60%)	Unaware 20(40%)
BUYERS AWARE OF PARASITES IN WATER	Aware 23(46%)	Unaware 27(54%)
DO BUYERS WASH THE PRODUCE BEFORE SELLING TO CONSUMERS	Yes 37(74%)	No 13(26%)
DO FARMERS USE THE SAME WATER FOR REFRESHING AFTER WORK	Yes 50(100%)	No 0 (0%)