

Diversity, Abundance and Distribution of Parasites of Medical Importance in Surface Water: A Case Study of Adada River, Enugu State, Nigeria

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Abstract Adada River, an all-the-year-round sparkling-clear river, and a very useful surface water was evaluated for parasites of medical importance, their diversity, abundance and distribution as a qualitative microbial risk-assessment (QMRA) factor. Parasites were enumerated by Stoll's Counting Method and identified by the morphological characteristics of their potentials. 14 genera of parasites were detected in the dry season, and 13 detected in the rainy season. Ten of these were potential human pathogens (*Taenia* spp, *Entamoeba histolytica*, *Schistosoma mansoni*, *S. haematobium*, *Ascaris lumbricoides*, *Giardia lamblia*, Hookworm, *Trichuris trichiura*, *Strongyloides stercoralis* and *Enterobius vermicularis*). Lowest average parasite per milliliter in the stations was 7.0×10^3 , and highest was 2.2×10^4 /ml. The most frequently encountered parasites were potential human pathogens (*Taenia* spp, *G. lamblia*, *E. histolytica*, hookworm and *S. mansoni*). Variation in the parasites' genera between the two seasons was not statistically significant ($p < 0.05$). In conclusion, Adada River is not potable and suitable for recreation, grazing and agriculture. All-the-year-round sparkling-clear surface water, such as this, may be a health deceit. In QMRA, parasites of medical importance should be a paradigm; likewise, in environmental microbiology, geographical coordinates of sample sites rather than physical landmarks, should be a paradigm, for better follow-ups.

Keywords: parasites, river, diversity, abundance, distribution, Stoll's counting technique

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1. Introduction

According to Amadi and Onyemelukwe [1], water is the common name assigned to the liquid state of hydrogen-oxygen compound, with the molecular formula H_2O , chemical structure of H-O-H, and IUPAC name of hydrogen/hydroxonium ion (depending on the oxidation state). Parasitism, is a phenomenon by which certain creatures, known as parasites, choose as a way of life, to live inside or attached to other organisms described as host, whereby they derive benefits such as nutrition, transportation, accommodation, protection, etc., with or without reciprocal benefit to the host. They become a problem when their association with the hosts constitutes a menace called diseases; hence they are grave source of concern to humanity, demanding serious attention. Water itself is the most important element in the universe and preventing of water pollution is vital for the health of

creature survival FeizHadad *et al.* [2]; its importance among all creations on earth is second only to air [3].

Parasitic diseases are among the most important causes of morbidity and mortality in developing countries [4]. Studies in different parts of the world by many scientists [5-10] indicated that water is the main source of parasites transmission and epidemics. Within these concepts of parasites and water, waterborne parasite infections are considered a re-emerging threat [11], Rosado-Garcia *et al.* [12] also noted that health systems, sanitation and water access have certain limitations in nations of Latin America: typical matters of developing countries. Nigeria and Africa, like the entire Third World nations, belong to this classification too. This is because water is often contaminated and therefore unhealthy for the consumers and users, and information on prevalence and detection of waterborne parasites in Third-World countries, particularly in rural areas is poor. Besides, it has been widely emphasized that there is paucity of simple coherent methodology for detection of parasites of medical

importance in surface water (in areas where DNA sequencing is not feasible or affordable) despite their high vulnerability to extreme weather events related to waterborne infections. So also, has only few reports been documented in this field in Nigeria. Addition to this misery is that waterborne diseases occur worldwide, and outbreaks caused by the contamination of community water systems have the potential to cause disease in large numbers of consumers. It will be more succinctly understood if specifically pointed out that waterborne outbreaks have economic consequences beyond the cost of health care for affected patients, their families and contacts, plus the economic costs and lost from bearing the ailment and disease. This is because they also create lack of confidence in potable water quality and in the water industry in general; for instance, many people do not drink tap water, including this author. Therefore, topic of emerging pathogens and related waterborne diseases should be considered a worldwide problem not to be restricted to sovereignty because human activities are expanding and travel between different countries and nations are increasing, for both tourism and business. Likewise, detection of such emerging or re-emerging waterborne parasites need dissemination of the information to increase awareness of newly characterized potential parasites or re-emerging parasites to appropriate people and authorities in the medical, public health and water and food communities.

Besides, many professional bodies have given selective attention to various sorts of health hazards; for example, the Royal Society of Tropical Medicine and Hygiene (RSTMH) has as one of its pet's project, what they call "Neglected Tropical Diseases" (i.e. leprosy, onchocerciasis, trachoma, lymphatic filariasis, schistosomiasis and soil transmitted helminths); the World Health Organization (WHO) has what it classifies as "Tropical Diseases" (i.e. African trypanosomiasis, malaria, schistosomiasis, onchocerciasis, leishmaniasis, lymphatic filariasis, Chagas disease and Dengue fever). These pet diseases were chosen in spite of apparent fear that other parasites of medical importance, such hookworm, *Ascaris lumbricoides*, *Giardia lamblia*, *Entamoeba histolytica*, *Strongyloides stercoralis*, *Enterobius vermicularis*, etc. are being neglected, and despite that they are all still prevalent and transmissible in water bodies, particularly in Third-World countries. For instance, Plate 1 is photo of about 100 *Ascaris lumbricoides* worms surgically removed from a 7 years-old female child in a University Teaching hospital in Enugu, Nigeria in October 2013 after intestinal occlusion.

Further, many times in history, apparently sparkling-clear water killed humans and animals after being drunk. This had led to many fatalities when they are consumed raw, untreated. This is especially the case with apparently sparkling-clear natural bodies of water (rivers, lakes, dams, etc.) that had led to mass killing of animals and humans alike after consumption. Where such cases had been most reported are among soldiers and refugees during wars, and wandering hunters in normal situations; in community settlements, it happens as water intoxications or water-borne diseases of various kinds.

This brought forth an all-the-year-round sparkling-clear Adada River in Enugu State, Eastern Nigeria which is the source of water for domestic, industrials, agricultural and

commercial purpose for more than nineteen towns and villages located within and beyond two Local Government Areas (LGA). This water is fetched by tanker drivers and distributed farther and wider, untreated, and has never been analyzed. It is also the source of water for Fulani herdsmen and their cattle, a secondary schools and about-to-be-embarked proposed site for Adada Campus of Enugu State University of Science and Technology, as well as it is the site for an ongoing millions-dollar Adada Dam project. It is such an important source of community water as could be the case in many parts of the world.

To the best of knowledge and according to reports from literature there is no study for detection of parasitic contamination of this important water source as a way of qualitative microbial risk assessment (QMRA), and as a structured and heterogeneous system as was indicated for soil by Nannipieri *et al.* [13]. Therefore, this study aimed to determine the diversity, abundance and distribution of parasites of medical importance along the river flow with respect to surrounding vegetation and river use, using formal diagnostic methods which will indicate specific pictures.

The specific objective of this study were to (i) examine Adada River, as an all-the-year-round sparkling-clear river and a very useful surface water for possible parasites of medical importance, (ii) evaluate the diversity and abundance of the detected parasites as a human risk factor, (iii) to determine the parasites' distribution along the river flow with respect to some surrounding vegetation and river use and, (iv) to evaluate the seasonal (dry and rainy) variations.

2. Materials and Methods

2.1. Study Area

The Adada River is the study area and sampling site that ran through Uzo-Uwani- and Igbo-Etiti Local Government Areas of Enugu State in Eastern Nigeria, at approximately five kilometers North-West of Aku, a village located 6°40' N and 7°18' E on the geographical map (6°42'7"N and 7°19'56"E on "Infinix Hot 7" smart phone-compass, measured at the Post Office). It is also the original site of the proposed Adada Campus of Enugu State University of Science and Technology as well as the location of the defunct Federal Government of Nigeria military cadet training school in the 1960s. Presently, it is the site of the on-going 2.8 billion Naira (about \$6,842,105.26) "Adada Dam" instituted by the government of the Federal Republic of Nigeria in 2011

2.2. Sampling Sites/Stations

The sampling areas were selected according to the surrounding vegetation's cover and river use at six differently determined, measured, and specified geographical coordinates (Stations 1-6) as follows:

Station 1 is geographical coordinate: 6°42'2"N 7°17'19"E ("Infinix Hot 7" smart phone-compass). It was upstream, towards the water source where there is limited human activity; the vegetation was originally rainforest, but in the distant past slightly disturbed by water tanker

drivers that created a path to the river from where they were then fetching water they sold to the local communities.

Station 2 is geographical coordinate 6°44'20"N 7°16'50"E. I was ways downstream from station1, at the beginning of where the river water was diverted for an ongoing Adada River Dam construction; the vegetation is only still slightly virgin, and disturbed by Fulani herdsmen that occasionally graze cattle along the bank of the river, and it is the camping site of the construction workers.

Station 3 is geographical coordinate 6°44'25"N 7°16'49"E. It was about the foot of the embankment where the Adada River water was diverted for the ongoing construction of the dam, and heavily disturbed by the ongoing construction work, and tanker driver that come to fetch water they sell to the local communities and beyond.

Station 4 is geographical coordinate 6°44'17"N 7°16'37"E. It was down-stream, a bit from the tail of the dam proper where from far and wide there are human activities, such as washing of clothes, soaking of cassava for fermentation, swimming, picnics, farmland at both banks, and point where Fulani herdsmen occasionally bring their cattle to drink water.

Station 5 is geographical coordinate 6°44'13"N 7°16'32"E. It was the temporary run-off point downstream for the diverted water flow from the dam, and also heavily disturbed on both banks of the river by heavy human activities, such as damming, farmlands, etc.

Station 6 is geographical coordinate 6°44'11"N 7°16'29"E. It was a little way downstream from station 5, before a former animal husbandry established by Eastern Nigeria Development Corporation (ENDC/ADP), also where Adada Secondary School [old site of the defunct Adada Campus of Enugu State University of Science and Technology(ESUT)] students fetch water, bath, wash clothes, swim, fishing, etc.

2.3. Determination of Geographical Coordinates

The digital phone-compass App was downloaded and installed into the "Infinix Hotspot 7" smart phone from the internet. At the precise chosen location or spot, the smart phone was put on and the compass icon clicks on and

waited for the application to booth. As soon as the phone-compass App booths, it brings out the precise geographical coordinate of the spot, which was then read off and recorded.

2.4. Collection of Water for Analysis

At each sampling station, water samples were collected in duplicates at some distance from the shore with clean pre-sterilized 500-ml bottles with stoppers. The bottles were first aseptically opened five centimeters (5cm) below the water surface, rinsed with the first set of water samples, and then filled with the required water sample, and the bottle aseptically closed. These were done between 10.00am to 12.00pm (late morning to early afternoon by which human activities have resumed), and done in two different sampling periods, June 13, 2016 (rainy season) and February 27, 2017 (dry season), precisely at the geographical coordinates. The samples were transported to the laboratory under ice and stored at 0°C until they were ready for analysis. A total of 24 water samples were collected (6 stations x duplicates samples = 12 x 2 seasons = 24 total). Total of 26 sample analysis were done (24 water samples plus control x 2 seasons).

2.5. Isolation and Enumeration of Parasites

Parasites were initially isolated using a slightly adjusted Finch [14] method (but this was later discarded when it was discovered that the very careful methodical counting process was more adequate. The enumerations of parasites were by Stoll's Counting Technique as for parasites in fluid and watery specimen [15], except that normal saline were used in the dilutions.

2.6. Detection and Identification of Parasites

Parasites were microscopically detected and identified in each water samples by their various morphological properties (as molecular methods could not have discerned), through their potentials (ova, cysts, larvae, oocyst and adults) [15]. The procedure is as follows in Table 1:

Table 1. Summary of the descriptive identification procedures for the isolated parasites in Adada River

Parasites	Descriptive identification
<i>S. stercoralis</i>	Microscopically identified as large, unshathed, active mobile rhabditiform larva, measuring about 250µm x 16µm, showing characteristics large bulbous oesophagus, differentiated from hookworm larvae by shorter mouth cavity
Hookworms (identified as ova and larvae)	Ova, which are colourless, thin-shell (which appear as black line around an ovum), oval in shape and about 65 x 40µm in diameter, usually segmented with 4-8 cell-stage, and distinguished from the ova of <i>Trichostrongylus</i> spp, <i>Ternidens deminitus</i> , <i>S. fuelleborni</i> and <i>Oesophagostom</i> spp; and Larvae, distinguished from <i>S. stercoralis</i> larvae by its characteristic deeper buccal cavity.
<i>T. trichiura</i>	Identified by a characteristic yellow-brown, barrel-shaped ovum, about 25–50µm in size with colourless protruding mucoid plug at each end.
<i>A. lumbricoides</i>	Identified by decorticated, fertilized and unfertilized eggs: fertilized eggs that were about 50–70µm x 30–50µm in length and breath, respectively, yellow-brown in colour, oval in shape, and containing a central granular mass covered by a shell with uneven albuminous coat; unfertilized eggs which were darker in colour and contains a central mass with larger granules that is covered by a thinner wall with more albuminous coat, and more elongated (90 x 45µm in size) than the fertilized one
<i>S. mansoni</i>	Identified by eggs that were oval in shape, pale yellow-brown in colour, and measuring about 60 – 150µm with, at times, fully visualizable internal fully developed miracidium, and with the characteristic single lateral spine.
<i>S. haematobium</i>	Identified by large eggs (145-45µm in length and breath, respectively), that are pale yellow-brown in colour and oval in shape, each containing a fully developed miracidium and the characteristic single terminal spine.

Parasites	Descriptive identification
<i>Taenia</i> spp	Identified by round eggs of about 30–40µm in diameter, containing barely visible oncosphere that is surrounded by thick, brown radially striated wall.
<i>E. vermicularis</i>	identified by colourless eggs measuring about 30–50µm that were oval in shape and flattened on one side, and containing barely visualizable larva.
<i>E. histolytica</i> (identified by trophozoites and cysts)	Trophozoites with active unidirectional amoeboid movement, unit nucleus that has barely discernible central karyosome, and measuring about 20–25µm in size; and Cysts that were round (10–15µm in diameter), containing 1–4 nuclei with barely discernible central karyosome, and having some chromatoid bodies in immature ones, and distinguished from larger <i>E. coli</i> (15–30µm in size), with 1–8 nuclei, and at times needle-like chromatoid body.
<i>I. butschlii</i>	Identified by small cysts (9–15µm in sizes) with only one nucleus that has compact mass of glycogen inclusion and no chromatoid body Trophozoites were small, pear-shaped flagellates (12–15 x 5–9µm in length and breadth, respectively) with rapid tumbling and spinning motions, having some discernible structures like four pairs of flagella, two axonemes, two discernible nuclei, large concave sucking disc located on the ventral surface, one or two curved median bodies; and by
<i>G. lamblia</i> (identified by its trophozoites and cysts)	Cysts that are also very small (7–12µm in diameter) with some discernible internal structures in the saline medium (e.g. four nuclei, remain of flagella, axonemes and median body). Both cysts and trophozoites were carefully differentiated from those of other flagellates of medical importance: <i>C. mesnili</i> , <i>Retortamonas intestinalis</i> , <i>E. hominis</i> and <i>Pentatrichomonas hominis</i> by their trophozoites with single nucleus, fewer flagella, shapes and smaller sized cysts that have not the characteristic appearances of <i>G. lamblia</i> (i.e. remains of flagella, four nuclei grouped to one end, and peculiar shape).
<i>C. mesnili</i>	Identified by lemon-shaped cysts that are smaller in size (<8µm) than those of other medical important flagellates (<i>R. intestinalis</i> = pear-shaped), and containing no remains of internal structure like <i>G. lamblia</i> .
<i>B. coli</i> (identified by trophozoites and cysts)	Trophozoites seen as large ciliates (50–200µm x 40–70µm in length and breadth, respectively), with rapidly revolving movement, well discernible macro-nucleus, two contractile vacuoles, discernible cilia beating at the region of the funnel-shaped cytostome when carefully focused; and by the round and thick-walled cysts that are also large (50–60µm in diameter) with discernible cilia lining the wall of the cyst
<i>B. hominis</i>	Identified as small round protozoa (about 15–30µm in size), with peripheral cytoplasm, a central vacuole, no discernible nucleus even at x40, and a granule which form a ring around the periphery

2.7. Statistical Analysis

The results were statistically analyzed with Students t-test ($p < 0.05$). Further, it was used to determine if the significant difference between the rainy- and dry season results, as well as the significant difference within the result obtained among the various stations ($p < 0.05$). Differences were analyzed through Chi-square and a P value of < 0.05 as significant.

3. Results

3.1. Diversity of Parasites of Medical Importance

The diversity and abundance of parasites of medical importance detected were more common than imagined. A total of 15 genera were detected in both the dry and rainy seasons as follows: *Taenia*, *Giardia*, *Entamoeba*, Hookworm, *Schistosoma*, *Ascaris*, *Endolimax*, *Balantidium*, *Iodamoeba*, *Hymenolepsis*, *Chilomastix*, *Strongyloides*, *Trichuris*, *Blastocystis* and *Enterobius* [Tables 2(Dry season) and 3 (Rainy season)]. Except *Enterobius*, all the above genera were detected in dry season, giving it a total of 14 types of genera. Likewise, except *Endolimax* and *Hymenolepsis*, all the above genera were detected in rainy season, giving it a total of 13 types of genera. Table 4 show the number of the types of genera found in each stations (and their duplicates) from the river water samples. However, there is no statistically significant difference ($p < 0.05$) in the number of the types of genera found in either the stations or its duplicate water samples between the rainy and dry seasons. In general, 11 of these detected parasites were potential pathogens (*Taenia* spp, *G. lamblia*, *E. histolytica*, Hookworm, *S. haematobium*, *S. mansoni*, *A. lumbricoides*,

B. coli, *S. stercoralis*, *T. trichiura* and *E. vermicularis*). Miracidia and cercaria were also detected in the dry season, but could not be classified as either *S. mansoni* or *S. haematobium*; non was detected in the rainy season.

3.2. Distribution of Parasites of Medical Importance

In the dry season (Table 2), Stations 3 and 4 (downstream) has the highest abundance of parasites per ml, while Stations 1 and 2 (upstream) the least abundant, unlike what was obtained in rainy season. Also, unlike what was obtained in the rainy season, distribution of parasites rather increases downstream. Similarly, from Table 4, there was no significant difference in the distribution of the types of genera throughout the stations ($p < 0.05$).

In the rainy season (Table 3), Stations 1 and 2 (upstream) have highest abundance of parasites per ml, while Station 5 (downstream) has the least. The picture therefore is that the distribution of parasites surprisingly decreases downstream in the rainy season, reflecting river use and vegetation as a factor. Rainy season has significantly ($p < 0.05$) higher abundance of parasites (1.6250×10^4 /ml) than the dry season (1.0958×10^4 /ml) as calculable from Table 2 and Table 3. From the result in Table 4, there was no significant difference in distribution of the types of genera throughout the 6 stations ($p < 0.05$).

3.3. Abundance of Parasites of Medical Importance

Table 5 show the dry and rainy season's abundance of the parasites in the 6 stations put together. Dry season 7 most abundant parasites, in descending order, were as follows: *Taenia* sp, *G. lamblia*, *E. histolytica*, *E. coli*. Hookworm, *S. haematobium*, and *S. mansoni*. Rainy

season 8 most abundant parasites almost followed the same sequence, in descending order are as follows: *E. histolytica*, *Taenia* spp, Hookworm, *G. lamblia*, *S. mansoni*, *E. coli*/*I. butschlii* and *A. lumbricooides* (Table 5). These also showed that most of the detected parasites were potential pathogens of man. The 3 least detected parasites in the dry season were: *S. stercoralis*, *T. trichiura* and *B. hominis*; the 3 least detected in the rainy season were: *C. mesnili*, *T. trichiura* and *E. vermicularis*. From Tables 5, if each abundance of the parasites is divided by 12(6 duplicate stations), it will give the average concentrations of each parasites per milliliter

of water sample. For instance, the average concentration of *Taenia* spp is $33500/12 = 2791.7$ per ml. Likewise from Table 2, the average number of parasites per milliliter of water sample in the dry season is: $131500/12 = 10,958$ (1.0958×10^4) parasites per ml. Likewise, from Table 3, the average concentration of parasites during the rainy season was: $195000/12 = 16,250$ (1.625×10^4) parasites per milliliter of water sample. Therefore, the concentration of parasites in the river was higher in the rainy season than the dry season. Lastly, Miracidia and cercaria were also detected at average concentration of $3000/12 = 250$ (2.5×10^2) per ml in the dry season.

Table 2. Adada River Parasites of Medical Importance: Diversity, Abundance and Distribution in Dry Season

	Stations	1A	1B	2A	2B	3A	3B	4A	4B	5A	5B	6A	6B
Diversity (/ml)	Abundance/%												
<i>Taenia</i> sp	$3.35 \times 10^4/25.5\%$	5.0×10^2	2.5×10^3	1.5×10^3	1.5×10^3	4.0×10^3	4.5×10^3	0	2.5×10^3	1.0×10^3	2.5×10^3	4.0×10^3	9.0×10^3
<i>Entamoeba. coli</i>	$1.25 \times 10^4/9.5\%$	5.0×10^2	0	0	0	5.0×10^2	0	5.0×10^3	1.0×10^3	3.0×10^3	0	2.0×10^3	5.0×10^2
<i>E. histolytica</i>	$1.7 \times 10^4/12.9\%$	1.0×10^3	0	0	0	4.5×10^3	0	3.5×10^3	5.0×10^2	2.0×10^3	0	5.5×10^3	0
<i>B. coli</i>	$2.5 \times 10^3/1.9\%$	0	0	0	0	0	0	0	0	0	0	2.5×10^3	0
Cercaria/Miracidia	$3.0 \times 10^3/2.3\%$	0	0	0	0	0	0	0	0	5.0×10^2	0	2.0×10^3	5.0×10^2
<i>S. mansoni</i>	$6.5 \times 10^3/5.1\%$	0	1.0×10^3			5.0×10^2	5.0×10^2	1.5×10^3	5.0×10^2	1.0×10^3	0	5.0×10^2	1.0×10^3
<i>S. haematobium</i>	$7.0 \times 10^3/5.3\%$	0	1.5×10^3	5.0×10^2	3.0×10^3	0	0	5.0×10^2	0	5.0×10^2	1.0×10^3	0	0
<i>A. lumbricooides</i>	$5.0 \times 10^3/3.8\%$	5.0×10^2	1.0×10^3	0	0	0	5.0×10^2	0	0	0	1.5×10^3	0	1.5×10^3
<i>Giardia lamblia</i>	$2.3 \times 10^4/17.5\%$	0	0	1.0×10^3	0	4.0×10^3	3.5×10^3	0	2.5×10^3	1.5×10^3	3.0×10^3	6.0×10^3	1.5×10^3
Hookworm	$1.0 \times 10^4/7.6\%$	0	5.0×10^2	5.0×10^3	5.0×10^2	0	1.0×10^3	5.0×10^2	1.0×10^3	5.0×10^2	0	5.0×10^2	5.0×10^2
<i>T. trichiura</i>	$5.0 \times 10^2/0.4\%$	0	0	0	0	0	0	0	0	0	0	5.0×10^2	0
<i>S. stercoralis</i>	$5.0 \times 10^2/0.4\%$	0	0	5.0×10^2	0	0	0	0	0	0	0	0	0
<i>I. butschlii</i>	$2.0 \times 10^3/1.5\%$	0	0	0	5.0×10^2	1.0×10^3	0	0	0	0	0	5.0×10^2	0
<i>C. mesnili</i>	$1.0 \times 10^3/0.8\%$	0	0	0	0	0	1.0×10^3	0	0	0	0	0	0
<i>E. nana</i>	$5.0 \times 10^3/3.8\%$	0	5.0×10^3	0	0	0	0	0	0	0	0	0	0
<i>B. hominis</i>	$5.0 \times 10^2/0.4\%$	0	0	0	0	0	0	0	0	0	0	5.0×10^2	0
<i>H. diminuta</i>	$2.0 \times 10^3/1.5\%$	0	0	0	0	0	0	0	0	0	0	0	2.0×10^3
Total counted	1.315×10^5	2.5×10^3	1.15×10^4	8.5×10^4	5.5×10^3	1.45×10^4	1.1×10^4	1.1×10^4	8.0×10^3	1.0×10^4	8.0×10^3	2.45×10^4	1.65×10^4
Total/Mean in stations	6.575×10^4	7.0×10^3		7.0×10^3		1.275×10^4		9.5×10^3		9.0×10^3		20.5×10^4	

Key: 1A to 6B= Station.

Table 3. Adada River Parasites of Medical Importance: Diversity, Abundance and Distribution in Rainy Season

	Stations	1A	1B	2A	2B	3A	3B	4A	4B	5A	5B	6A	6B
Diversity (/ml)	Abundance/%												
<i>Taenia</i> sp	$4.5 \times 10^4/23.1\%$	1.0×10^3	8.0×10^3	1.0×10^3	1.0×10^3	1.0×10^4	4.0×10^3	6.0×10^3	2.0×10^3	1.0×10^3	2.0×10^3	4.0×10^3	5.0×10^3
<i>Entamoeba. coli</i>	$6.0 \times 10^3/3.1\%$	0	3.0×10^3	1.0×10^3	0	0	1.0×10^3	0	0	0	0	0	1.0×10^3
<i>E. histolytica</i>	$6.6 \times 10^4/33.9\%$	1.1×10^4	8.0×10^3	5.0×10^3	1.4×10^4	0	1.5×10^4	2.0×10^3	2.0×10^3	3.0×10^3	2.0×10^3	4.0×10^3	0
<i>Balantidium coli</i>	$2.0 \times 10^3/1.0\%$	2.0×10^3	0	0	0	0	0	0	0	0	0	0	0
<i>Schistosoma. mansoni</i>	$1.1 \times 10^4/5.6\%$	1.0×10^3	1.0×10^3	0	0	0	0	1.0×10^3	1.0×10^3	1.0×10^3	0	4.0×10^3	2.0×10^3
<i>Ascaris. lumbricooides</i>	$5.0 \times 10^3/2.6\%$	0	1.0×10^3	1.0×10^3	1.0×10^3	0	0	0	0	2.0×10^3	0	0	0
<i>Giardia</i> sp	$2.3 \times 10^4/11.8\%$	0	2.0×10^3	2.0×10^3	2.0×10^3	0	3.0×10^3	0	5.0×10^3	5.0×10^3	1.0×10^3	2.0×10^3	1.0×10^3
Hookworm	$2.5 \times 10^4/12.8\%$	0	0	2.0×10^3	11000	4.0×10^3	2.0×10^3	1.0×10^3	2.0×10^3	1.0×10^3	0	2.0×10^3	0
<i>Trichuris trichiura</i>	$1.0 \times 10^3/0.5\%$	0	0	0	0	0	0	0	1.0×10^3	0	0	0	0
<i>Enterobius. vermicularis</i>	$1.0 \times 10^3/0.5\%$	0	0	0	0	0	0	0	0	0	0	1.0×10^3	0
<i>Strongyloides. stercoralis</i>	$3.0 \times 10^3/1.5\%$	0	0	0	0	0	0	0	0	0	0	0	3.0×10^3
<i>Iodamoeba butschlii</i>	$6.0 \times 10^3/3.1\%$	0	6.0×10^3	0	0	0	0	0	0	0	0	0	0
<i>Chilomastix. mesnili</i>	$1.0 \times 10^3/0.5\%$	0	0	0	0	1.0×10^3	0	0	0	0	0	0	0
Total counted	1.95×10^5	1.5×10^4	2.9×10^4	1.2×10^4	$.9 \times 10^4$	1.5×10^4	2.5×10^4	1.0×10^4	1.3×10^4	1.3×10^4	5.0×10^3	1.7×10^4	1.2×10^4
Total/Mean in stations/ml	9.75×10^4	2.2×10^4		2.05×10^4		2.0×10^4		1.15×10^4		9.0×10^3		1.45×10^4	

Table 4. Genera and parasitic load in the Adada River at various stations in the dry and rainy seasons

Stations/Variable	SEASON												
	Dry season	1A	1B	2A	2B	3A	3B	4A	4B	5A	5B	6A	6B
Number of genera in duplicate stations		4	6	5	4	6	6	5	6	8	4	11	8
Number of types of genera		8		6		9		7		9		13	
Number of parasites in stations		1.4 x 10 ⁴		1.4 x 10 ⁴		2.55 x 10 ⁴		1.9 x 10 ⁴		1.8 x 10 ⁴		4.1 x 10 ⁴	
Average parasites per station		7.0 x 10 ³		7.0 x 10 ³		1.275 x 10 ⁴		9.5 x 10 ³		9.0 x 10 ³		2.05 x 10 ⁴	
	Rain Season	1A	1B	2A	2B	3A	3B	4A	4B	5A	5B	6A	6B
Number of genera in duplicate stations		4	7	6	5	3	5	4	6	3	6	5	
Number of types of genera		8		6		6		6		6		8	
Number of parasites in stations		4.4 x 10 ⁴		4.1 x 10 ⁴		4.0 x 10 ⁴		2.3 x 10 ⁴		1.8 x 10 ⁴		2.9 x 10 ⁴	
Average parasites per station		2.2 x 10 ⁴		2.05 x 10 ⁴		2.0 x 10 ⁴		1.15 x 10 ⁴		9.0 x 10 ³		1.45 x 10 ⁴	

Table 5. Percentage abundance and prevalence of parasites in Adada River in rainy & dry seasons

Parasites	Abundance/% (Dry season)	Ranking	Abundance/% (Rainy season)	Ranking
<i>Taenia</i> spp	3.35 x 10 ⁴ /25.5%	1 st	4.5 x 10 ⁴ /23.1%	2 nd
<i>Giardia lamblia</i>	2.3 x 10 ⁴ /17.5%	2 nd	2.3 x 10 ⁴ /11.8%	4 th
<i>Entamoeba. histolytica</i>	1.7 x 10 ⁴ /12.9%	3 rd	6.6 x 10 ⁴ /33.9%	1 st
<i>Entamoeba. coli</i>	1.25 x 10 ⁴ /9.5%	4 th	6.0 x 10 ³ /3.1%	6 th
Hookworm	1.0 x 10 ⁴ /7.6%	5 th	2.5 x 10 ⁴ /12.8%	3 rd
<i>Schistosoma. haematobium</i>	7.0 x 10 ³ /5.3%	6 th	0	-
<i>Schistosoma. mansoni</i>	6.5 x 10 ³ /5.1%	7 th	1.10 x 10 ⁴ /5.6%	5 th
<i>Ascaris lumbricoides</i>	5.0 x 10 ³ /3.8%	8 th	5.0 x 10 ³ /2.6%	7 th
<i>Endolimax. nana</i>	5.0 x 10 ³ /3.8%	8 th	0	-
Cercaria/miracidia	3.0 x 10 ³ /2.9%	9 th	0	-
<i>Balantidium. coli</i>	2.5 x 10 ³ /1.9%	10 th	2.0 x 10 ³ /1.0%	9 th
<i>Iodamoeba butschlii</i>	2.0 x 10 ³ /1.5%	11 th	6.0 x 10 ³ /3.1%	6 th
<i>Hymenolepis. diminuta</i>	2.0 x 10 ³ /1.5%	11 th	0	-
<i>Chilomastix. mesnili</i>	1.0 x 10 ³ /0.8%	12 th	1.0 x 10 ³ /0.5%	10 th
<i>Strongyloides. stercoralis</i>	5.0 x 10 ² /0.4%	13 th	3.0 x 10 ³ /1.5%	8 th
<i>Trichuris. trichiura</i>	5.0 10 ² /0.4%	13 th	1.0 x 10 ³ /0.5%	10 th
<i>Blastocystis. hominis</i>	5.0 10 ² /0.4%	13 th	0	-
<i>Enterobius. vermicularis</i>	0	-0	1.0 x 10 ³ /0.5%	10 th

4. Discussion

According to Sures *et al.* [16] parasites are attracting increasing interest from parasite ecologists as potential indicators of environmental quality because of the variety of ways in which they respond to anthropogenic pollution. The high diversity of parasites of medical importance in the river water should be something of very serious concern, especially since 11 of them were potential pathogens (*Taenia* spp, *G. lamblia*, *E. histolytica*, Hookworm, *S. haematobium*, *S. mansoni*, *A. lumbricoides*, *B. coli*, *S. stercoralis*, *T. trichiura* and *E. vermicularis*). This river water, as are found in many rural settings in the world (where about 65% of the world population resides) is drunk untreated. Detected parasites such as *G. lamblia*, *E. histolytica*, *A. lumbricoides*, *B. coli*, *T. trichiura* and *E. vermicularis* that has oral mode of transmissions make it a very serious source of health hazards. They are all serious pathogens of various sorts of

gastrointestinal diseases. *Giardia lamblia* and *Entamoeba histolytica* have been identified as significant waterborne pathogens and have been found responsible for several serious outbreaks worldwide over the past ten years [17,18]. *A. lumbricoides*, among many other things, can cause intestinal occlusion that calls for surgical emergency [19]. *T. trichiura* is an agent of the syndrome called anal prolapsed, or pile, or haemorrhoid [19,20]. *E. vermicularis* is a dangerous agent of very intense anal itch which has its most pronounced effect, such as psychiatry in infected infants and neonates.

Also, the river and what they could be their tastes in rural communities in Africa and the Third World, are used for agriculture, grazing and recreational values such as swimming, picnics, and clothes-washing; and it is a site for an on-going Adada Dam project. Men and cattle wade into it for other values such as drinking; fetching water, grazing, soaking cassava, fishing, the list is endless. When detected parasites such as Hookworm, *S. stercoralis*,

S. mansoni and *S. haematobium*, whose mode of transmission are active penetration of intact skin are considered in this river, the potential high risk of health hazards cannot be overestimated. The four are agents of various serious diseases with very poor prognosis if untreated [21,22]. In contrast to the work of Robertson and Gjerde [23], indeed, parasites of medical importance should be a paradigm in qualitative microbial risk assessment (QMRA). That *Cryptosporidium* sp was not detected in this study may likely be in accordance that it is not a prevailing problem in this part of the community.

High abundance of parasites per station ($9.0 \times 10^3 - 2.20 \times 10^4$) per ml (rainy season) and $7.0 \times 10^3 - 2.05 \times 10^4$ per ml (dry season)], as shown in Tables 3, was not a surprise because similar high amount of microbes had been demonstrated per ml, {though of soil sample [24-26]; but then, these are ultimately washed into surrounding surface waters}. As was extrapolated from Tables 2 and 3, added up together, the high average detected in dry (1.0958×10^4 /ml) and rainy (1.6250×10^4 /ml) seasons were, however, rather of concern. These were also demonstrated among the seven highest pathogens detected in the Adada River (Table 5). For instance, the concentration of *Taenia* spp was $33500/12 = 2792$ (2.792×10^3)/ml. That was one of the greatest concerns in this work because all the pathogenic parasites were detected in high concentration that could be of infective dosages. Infective dosage for *Cryptosporidium* oocyst has been found as low as 130 and that of *G. lamblia* as 50 – 100 cysts [1, 27]. What was obtained for *G. lamblia* in this work was $23000/12 = 1917$ /ml (dry season), same for rainy season. Hence, multiples of the milliliters of Adada River water normally drunk could then give imagination of the potential health hazards being undergone for so long. Hamid and Siddiqui [28] stated similar results from Columbia University in three groups of population that used water from deep wells, protected springs and surface water which showed *Giardia* and *Entamoeba histolytica* were the most common in second and third groups.

Such concern should not be limited to the findings in this work in Nigeria, because various researchers had reported that waterborne diseases occur worldwide, and outbreaks caused by the contamination of community water systems have the potential to cause disease in large numbers of consumers [12], but where this is of most worry is that most of them are not documented or highlighted in Third World countries where more than 65% of the world population reside. Interesting too, was the fact that in spite of such abundance parasites, *Escherichia coli* was not detected in this river in a research work in 2014 [3].

In line with this study, there have been lots of reports that water play very important role in the distribution of many parasites which includes *G. lamblia* and *E. histolytica* [6]. It was not a surprise that rainy season has higher abundance of parasites than the dry season because river water during this season receives high amount of debris, which usually includes parasites from sewerages and run-offs. Where this is of most interest is that the sources of these parasites are from faeces defecated unto open soils by the surrounding communities through their unhealthy habits. The detected parasites reflect the prevailing pathogens from the neighboring

communities. Like this study, different studies have shown assorted prevalence rates, especially from various types of water sources; 16.6% for *Entamoeba histolytica/dispar* [29]; 2.2% *Entamoeba histolytica*, 6.6% *Giardia*, 1.7% *Ascaris* and 1.5% for *Hymenolepis nana* [29].

Why distribution of parasites decrease downstream during rainy season must have been because upstream vegetation provides more seclusions during this period which give room to hidings where the users of the river and the surrounding communities defecate onto the open soil from where the contents (including parasites) are washed into the river; whereas during the dry season less of such soiling on the bank of the river upstream is done and the river pollution almost strictly depends on the active use in this period. Further, that there was no statistical significant difference in the number of the types of genera in any of the station indicates that vegetation and river use do not greatly reflect the distribution of parasites of medical importance, which actually does not thrive or grow or multiply in surface water and are transient tenants depending on spatial input by available biotic sources. Although there have been many works on physico-chemical properties of water [30,31], for examples, no literature ever looked at the parasites index as was established in this work. There was consequently paucity of comparative analysis in that direction.

5. Conclusion

In conclusion, these results extend our knowledge on waterborne parasites of medical importance in a tropical surface water of a Third World country, and such occurrence information on waterborne pathogens assists the management of such water. The studies also underscore the need for adequate environmental management of such an important water resource. It as well calls for adequate parasites control programme and sanitation awareness in such untreated highly used river water.

6. Recommendation

Parasites of medical importance should be a paradigm in microbial risk assessment factor. In addition to this recommendation, if similar water like Adada River is to be further drunk raw, they must, at least, first be boiled for at least 15 minutes; that temperature and time should be enough to kill any vegetative parasites and microbe by the present disinfection technique. Further, Bakir *et al.* [32] reported a case were no parasite was seen in an untreated dam water, this work as well call for similar future standard towards an ongoing dam project, like Adada River Dam. . Lastly, geographical coordinates should be a paradigm in environmental microbiology (Amadi, *et al.*, 2020); it greatly assists future follow-up of precise locations, and it is simple, cost-less, and does not requires special technical know-how or training.

Additional Information

There is no competing financial and non-financial interest.

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