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Bio-characterization of the infectious forms of intestinal protozoan and helminthes in the streams and wells of the municipality of Bafia (Cameroon) and relationship to environmental factors

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Abstract

Water is essential for all life but it can also be a source or vector of many diseases and therefore cause death. The objective of this work is to analyze the quality of water in the streams and wells of the Council of Bafia (Central Region) in relation to water quality. The organoleptic parameters are determined by a spectrophotometer which allows the measurement of organoleptic variables such as Suspended Matter (MES); Color; Turbidity and Ammoniacal nitrogen. The Kato-Katz technique is the World Health Organization (WHO) "gold standard" that is used to assess the prevalence and infection intensity of soil and water transmitted protozoan and helminths. The formol-ether-concentration method is commonly used for the diagnosis of infectious protozoan and helminths. Organoleptic variables vary in streams, with TSS with 95mg/L, Color higher with 295 Pt. Co, Turbidity and Ammonia with an average of 0.4 mg/L in streams. *Cryptosporidium* sp has an average of 10-300 oocysts, *Cyclospora* sp is 225 oocysts and *Balantidium* has cysts in the first outing. In the second outing *Cryptosporidium* sp has an average of 200 oocysts, *Cyclospora* sp is 20 oocysts in the streams. In the first outing, the larva of *Strongyloides* is 50 larva and 25 oocysts for *Diphyllobothrium* in the second outing, there were no eggs identified. In the first outing, the larva of *Strongyloides* is 50 larva and 25 oocysts for *Diphyllobothrium* in the second outing, there were no eggs identified. It is necessary for researchers and scientists to understand protozoa and helminths and develop necessary tools to detect and eradicate them from the environment. Proper diagnostic and treatment technologies should be adopted in water reuse schemes to stop the spread of protozoa and helminth eggs in the environment.

Keywords: Intestinal parasites; Streams; Well; Physico-chemistry; Bafia

1. Introduction

Water is essential for all life, but it can also be a source or vector of many diseases and therefore cause death. The relationship between humans and water is complex and varied. Water is necessary for economic development, but dams or irrigation networks can be harmful to health through the diseases they carry (Desjardin, 1997). Water only appeared late as a possible role in health. It was only identified as a factor in the transmission of disease in 1854 during a cholera epidemic in London. Gradually, proper access to drinking water has increased life expectancy, provided that hygiene conditions improve at the same time. Water-related diseases are very diverse in their relationship to water and the mode of transmission (WWAP, 2017). Water quality refers to the condition and characteristics of water that determine its suitability for various uses and the health of aquatic ecosystems. It encompasses the chemical, physical, and biological properties of water and the presence of contaminants and pollutants. Monitoring and maintaining good water quality is crucial for human health, ecosystem sustainability, and the overall well-being of communities. Water pollution refers

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to the contamination of water bodies, like lakes, rivers, and oceans, with harmful substances. This contamination can be caused by various human activities, including industrial discharges, agricultural runoff, and improper waste disposal. Water pollution can negatively impact ecosystems, human health, and overall water quality (Avcievala, S. 1991),

Intestinal parasites are mainly transmitted orally, following the ingestion of eggs or larvae present in contaminated water or food, or by contact with soiled surfaces, then by bringing hands to the mouth (Lair, 2018). The frequency of intestinal parasitism in humans has been highlighted for many years by doctors on Reunion Island and in reports from the Directorate of Health and Social Action. But the first statistical evaluation of this endemic was carried out in 1957 by a mission of the World Health Organization (WHO), whose study, however, only focused in specific polluted areas.

It revealed the existence of polyparasitism in almost all school-aged children and recommended a fight focused on sanitation and health education of the population (Desjardin (1997). The presence of parasites in water, whether intended for consumption or other uses, can lead to public health problems. The parasites most frequently found in water are protozoa such as *Giardia* and *Cryptosporidium*, which can cause gastrointestinal infections. Other pathogens such as bacteria (e.g., *E. coli*, *Salmonella*) and viruses can also contaminate water and pose health risks (Vashishat, Aarushi ;2023)

Intestinal parasites present in streams and wells are transmitted mainly orally, through the ingestion of contaminated water or food (fruits and vegetables) washed in this water (Mampuya). The eggs or larvae of parasites present in human or animal feces, and which end up in the water, can contaminate the water source and infect those who consume it or use it to wash food: The objective of this work is to analyze the quality of water in streams and wells in the Council of Bafia; Central Region in relation to water quality (Vashishat et Aarushi, 2023)

2. Materials and Methods

Bafia is a city in Cameroon located in the Centre Region, 120 km north of Yaoundé. Bafia is the capital of the Mbam-and-Inoubou Department. It has a population of approximately 55,700. It is the third largest city in the Centre Region after Yaoundé and Mbalmayo. Bafia is a city and community in Cameroon's Central Province region. It is the capital of the Mbam-et-Inoubou Department. It is located 120 km north of Yaoundé.

Most of the citizens of this region belong to either the Bafia or the Yambassa people. The office of mayor of Bafia was established in 1960. The former department of Mbam was the current department of Bam-and-Inoubou and Mbam-and-Kim. Since 1992, Bafia has been the department of Mbam-and-Inoubou. Bafia also has a sub-prefecture to administer the district of Bafia, a community that includes the territorial headwaters of this community. All central county administration is carried out in the city by departmental and district delegates

The city of Bafia has a teacher training college (ENIEG), a classical high school, a technical high school, a technical, industrial and commercial college (cetic of Kiki), a bilingual high school, and three secondary or second cycle colleges. The Bafia District Hospital is the city's main medical facility. It has many specialties such as general medicine, surgery, gynecology, odontostomatology, pediatrics, etc.

The town also has two bakeries, as well as shops and markets, bars, and taverns. Several restaurants operate in the town, offering a variety of dishes dominated by local dishes such as Kepen, Kibazi, Kidjane, Bitooso, etc. A two-star hotel with a nightclub and several other similar establishments, albeit smaller, offer accommodation to visitors. Bafia is accessible by urban and interurban roads. Approaching Yaoundé, many road transport companies use the 121 km long Bafia road from the Tongolo bus station (north of Yaoundé).

Coming from Douala, one can either go via Yaoundé (360 km), or via Bafang and Bangangté or Bafang and Bafoussam (approximately 390 km in each case). The Biamo airfield, very active during the first fifteen years following independence, is no longer able to accommodate single-engine (mono-engine) planes, twin-engine planes or helicopters due to urbanization.

The Nguen River is 4 km long and crosses the main Yaoundé-Bafoussam road. The houses are upstream, with toilets 5 to 10 meters away, which are used for laundry, convicts, and traditional rituals. Household waste is dumped a few meters from the yards and into the river. The downstream point is about 2 km away, with the presence of a cocoa field. The Bigna River is 5 km long and crosses the Nigama market, a sugarcane field, a pigsty, and garbage dumps near marshy areas. The houses are upstream; with toilets 2 meters away, and the water is used for laundry, convicts, and traditional rituals. Household waste is dumped a few meters from the yards and into the river. The downstream point is about 4 km away. with the presence of cocoa fields in the area (Figure 1). The research started from January 2024 till date.

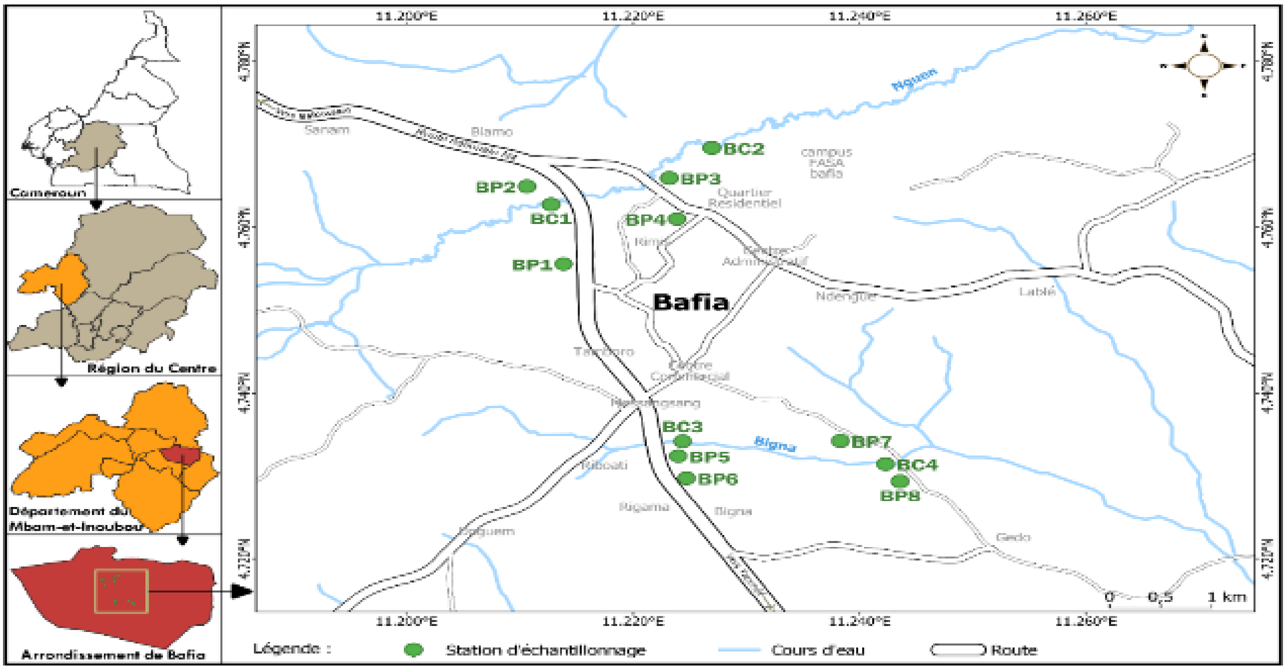

Figure 1 Study area, Municipality of Bafia

Organoleptic parameters are determined by a spectrophotometer that measures organoleptic variables such as TSS; Color; Turbidity and Ammoniacal Nitrogen. Observing ciliates involves looking at their cilia, the microscopic hair-like structures that allow them to move and feed. Counts are made under a microscope using a 40X objective; their ultrastructure and microphotographs are taken using a 100X objective.

The direct microscopic examination of feces is essential to detect parasitic elements such as the larvae of *Strongyloides stercoralis* which are motile. It is also usually adequate to detect high concentrations of the eggs of helminth infections with *Ascaris lumbricoides*. The main advantage of this method is that it is rapid and inexpensive. However, it is only semi-quantitative, and it is not often used in control programs. It is more widely used in the routine analysis to detect protozoan parasites such as the trophozoites of *Entamoeba histolytica, Giardia lamblia* and more rarely *Balantidium coli.* It involves emulsifying a small quantity of fresh stool in one drop of saline on a microscope glass slide. A thin smear preparation is obtained by placing a cover glass on the emulsified stool and examined under a light microscope to detect the eggs/larvae/trophozoites of parasite species. An eosin or iodine preparation is also necessary to identify the cysts/oocysts of intestinal protozoa.

The Kato-Katz technique is the WHO "gold standard" that is widely used to assess the prevalence and infection intensity of STHs. Amongst the copro-microscopic methods, Kato-Katz has several advantages including; high sensitivity, egg quantification, cost effectiveness and requires minimal infrastructure. It is possible to stratify infection intensities using egg counts and cut-off values. For the Kato-Katz technique, the centrifuged and sieved feces sample (approximately 41.7 mg, 20 mg, or 50 mg depending on the size of the template) is placed on a glass slide. The preparation is covered with a piece of cellophane soaked in glycerol. Subsequently, the slide is inverted and gently pressed down resulting in a thin smear. The added glycerol serves to 'clear' the fecal material (fat) from around the eggs. Hookworm eggs require about 30 min for this step, while for the other species, the reading of the slide under the microscope can be done after 1 to 24 h The eggs are then counted under the microscope and the count expressed in per gram of sedimented samples.

The formol-ether-concentration method is commonly is used for the diagnosis of infectious protozoan and helminths. The main advantage is that it is fast, and it allows for the concentration of a range of parasites. Both fresh and preserved samples can be used with this technique. The use of formol inactivates the organisms and thus minimizes the risk of laboratory-acquired infection from environmental pathogens. STHs as well as intestinal protozoa was diagnosed with this technique. When used in combination with the Kato-Katz method, the diagnostic sensitivity for helminths was greatly improved. The stool samples was fixed with either sodium acetate-acetic acid-formalin (SAF), or diluted formalin, to allow for sample storage and retrospective analyses. Several modifications of the technique have been made over the years. After adding ether or ethyl acetate, the mixed suspension is then centrifuged. The parasitic elements, cysts, oocysts, eggs, or larvae are fixed and sedimented, while the fecal debris are suspended in the layer between the ether and the formol water. The entire sediment is further examined under a light microscope to detect and count the parasite (Majaf, 2014).

3. Results and Discussion

Organoleptic variables vary in the streams, with higher MES in the Bigna with 95 mg L, higher color in the Ngyen 295Pt. Co, Turbidity in the Ngyen and Mok and ammonia with an average of 0.4 mg L in the streams (Table I-V)(Vashishat, Aarushi, 2023)

Table 1 Presentation of physicochemical parameters in watercourses

Stations	**BC1**	**BC2**	**BC3**	**BC4**
MES (mg/L)	12	6	95	22
Color (Pt.Co)	152	84	502	129
Turbidity (FTU)	22	12	144	24
Ammonia (mg/L)	0,17	0,06	0,64	0,25

Cryptosporidium sp has an average of 10-300 mg/L oocysts, *Cyclospora* sp is 225 mg/L oocysts and *Balantidium* has 75 mg/L cysts in the first outing. In the second outing *Cryptosporidium* sp has an average of 200 mg/L oocysts, *Cyclospora* sp is 20 mg/L oocysts in the streams.

Table 2a Protozoa in streams in Bafia (First outing)

Species/Station	**BC1**	**BC2**	**BC3**	**BC4**
Cryptosporidium sp	10	20	75	300
Isospora sp	0	0	0	0
Cyclospora sp	0	10	225	0
Sarcocystis sp	0	10	300	75
Giardia sp	0	0	0	0
Entamoeba histolytica	0	0	0	0
Entamoeba coli	0	0	0	0
Balantidium coli	0	0	75	0

Table 2b Protozoa in streams in Bafia (second outing)

Species /Station	**BC1**	**BC2**	**BC3**	**BC4**
Cryptosporidium sp	240	0	20	200
Isospora sp	0	20	0	0
Cyclospora sp	0	20	0	0
Sarcocystis sp	0	0	0	0
Giardia sp	0	0	0	0
Entamoeba histolytica	0	0	0	0
Entamoeba coli	0	0	0	0
Balantidium coli	0	0	0	0

Cryptosporidium sp has an average of 5-300 mg/L oocysts, *Cyclospora* sp is 100 mg/L oocysts and *Balantidium* has 60 mg/L cysts in the first outing. In the second outing *Cryptosporidium* sp has an average of 150 mg/L oocysts, *Cyclospora* sp is 100 mg/L oocysts in the streams

Table 3a Protozoa in wells in Bafia (first outing)

Species /Station	BP1	BP2	BP3	BP4	BP5	BP6	BP7	BP8
Cryptosporidium sp	16	20	0	0	36	300	5	0
Isospora sp	0	0	0	0	0	0	5	0
Cyclospora sp	0	20	0	0	30	100	0	0
Sarcocystis sp	16	0	0	0	24	0	10	0
Giardia sp	0	0	0	0	0	0	0	0
Entamoeba histolytica	0	0	0	0	0	0	0	0
Entamoeba coli	0	0	0	20	0	0	0	0
Balantidium coli	0	0	0	60	0	50	0	0

Table 3b Protozoa in wells in Bafia (Second outing)

Species /Station	BP1	BP2	BP3	BP4	BP5	BP6	BP7	BP8
Cryptosporidium sp	20	0	0	150	0	0	30	0
Isospora sp	0	0	0	0	0	0	10	0
Cyclospora sp	0	0	0	0	0	0	40	0
Sarcocystis sp	0	0	0	0	0	0	0	0
Giardia sp	0	0	0	0	0	0	0	0
Entamoeba histolytica	0	0	0	0	0	0	150	0
Entamoeba coli	0	0	0	0	0	0	0	0
Balantidium coli	0	0	0	0	0	0	0	0

In the streams the larva of *Strongyloides* is 257; *Ankysloma* has a value of 125 and *Ascaris* sp; *Trichostrongilus* eggs 75, and in the second outing; *Ankylostoma* is 200, the eggs of trichostogylus is 75, in the second outing the eggs of *Ankylostoma* is 200; *Enterobius* is 300, *Trichuris trichiura* 100,

Table 4a Helminth in Streams in Bafia (First outing)

Species /Station	BC1	BC2	BC3	BC4
Strongyloides larva	0	0	0	257
Ankylostoma eggs	0	0	0	125
Tenia sp eggs	0	0	0	0
Ascaris sp eggs	0	0	0	75
Faciola sp eggs	0	0	0	0
Hymenolepis sp eggs	0	0	0	0
Enterobus sp eggs	0	0	0	0
Trichostrongilus eggs	0	0	0	75

Diphyllobotrium eggs	0	0	0	0
Trichuris trichiura eggs	0	0	0	0
Schistosoma sp eggs	0	0	0	0

Table 4b Helminth in Streams in Bafia (second outing)

Species /Station	**BC1**	**BC2**	**BC3**	**BC4**
Strongyloides larva	0	0	20	0
Ankylostoma eggs	50	0	0	200
Tenia sp *eggs*	0	0	0	0
Ascaris sp *eggs*	0	0	0	75
Faciola sp *eggs*	0	0	0	0
Hymenolepis sp *eggs*	0	0	0	0
Enterobus sp *eggs*	300	0	0	0
Trichostrongilus eggs	0	0	0	75
Diphyllobotrium eggs	0	0	0	0
Trichuris trichiura eggs	0	0	0	100
Schistosoma sp *eggs*	0	0	0	0

In the first outing, the larva of *Strongyloides* is 50 larva and 25 oocysts for *Diphyllobothrium* in the second outing, there were no eggs identified.

Table 5a Helminth in streams in Bafia (First outing)

	MP1	**MP2**	**MP3**	**MP4**	**MP5**	**MP6**	**MP7**	**MP8**
Strongyloides larva	0	0	0	0	0	50	0	0
Enkylostoma eggs	0	0	0	0	0	0	0	0
Tenia sp *eggs*	0	0	0	0	0	0	0	0
Ascaris sp *eggs*	0	0	0	0	0	0	0	0
Faciola sp *eggs*	0	0	0	0	0	0	0	0
Hymenolepis sp *eggs*	0	0	0	0	0	0	0	0
Enterobus sp *eggs*	0	0	0	0	0	0	0	0
Trichostrongilus eggs	0	0	0	0	0	0	0	0
Diphyllobotrium eggs	0	0	0	0	0	25	0	0
Trichuris trichiura eggs	0	0	0	0	0	0	0	0
Schistosoma sp *eggs*	0	0	0	0	0	0	0	0

Table 5b Helminth in wells in Bafia (Second outing)

	BP1	BP2	BP3	BP4	BP5	BP6	BP7	BP8
Strongyloides larva	0	0	0	0	0	0	0	0
Ankylostoma eggs	0	0	0	0	0	0	0	0
Tenia sp *eggs*	0	0	0	0	0	0	0	0
Ascaris sp *eggs*	0	0	0	0	0	0	0	0
Faciola sp *eggs*	0	0	0	0	0	0	0	0
Hymenolepis sp *eggs*	0	0	0	0	0	0	0	0
Enterobus sp *eggs*	0	0	0	0	0	0	0	0
Trichostrongilus eggs	0	0	0	0	0	0	0	0
Diphyllobotrium eggs	0	0	0	0	0	0	0	0
Trichuris trichiura eggs	0	0	0	0	0	0	0	0
Schistosoma sp *eggs*	0	0	0	0	0	0	0	0

In many parasite–host systems, effective transmission of parasite offspring to new hosts is a critical component of parasite success (Tabe I-V). It is likely that tradeoffs between efficient use of host resources (conferring greater fitness costs to the host) and transmission of protozoa and helminth in the aquatic system is primordials for the infectious forms of parasites (Vashishat, Aarushi,2023). Parasite transmission may require close contact between host individuals as the children who bath in the streams, the population that uses wells as drinking water and the use of these streams for domestic chores. Parasites that grow and reproduce quickly by efficiently pillaging the host's body may render their hosts too ill to pass their progeny on to new hosts. An aggressive parasite could make a host too weak to move widely and interact with conspecifics, or could kill the host before any opportunity for transmission (Desjardin ,1997). This spread of infectious forms has important consequences for human disease conception. Many of the symptoms induced by parasites directly impacts the success of parasite transmission. It is likely no coincidence that the main symptoms of infections by waterborne parasites such *Giardia lamblia* that involves severe digestive distress, a surefire way to quickly reinfest water used by hosts (WHO,2018).

Some parasites may increase transmission success by adhering on organic matter, increasing the load of oocysts, cysts and eggs of infectious forms, low sanitary predictions as toilettes wastes are released into the water way. Healthier hosts may live longer, move further, and interact with more conspecifics, ultimately leading to greater transmission and dispersal for the parasite. Thus, in situations where transmission is limiting, natural selection may favor reduced virulence in the parasite populations. The enteric parasitic endemic is the consequence of the uncontrolled dispersal of human fecal matter on the soil. Depending on the nature of this soil, its physicochemical properties, climatic data (sunshine, humidity, etc.), the parasitic forms (cysts, eggs, larvae) contained in the stools will find favorable conditions for their development and resistance. They are the source of new contaminations, superinfections or reinfections after having soiled hands, water, vegetables (oral contamination) or by direct contact of humans with the soil or water (cutaneous penetration of the infective larvae of hookworms, eelworms). In addition to these direct contamination factors, there is the possibility of long-distance mechanical transmission of parasites present on the ground via flies, cockroaches, rodents, domestic animals or livestock (Golvan, 1990). The telluric and bioclimatic conditions of Reunion Island are favorable to the development of most parasitic species. The underdevelopment of sanitary equipment in terms of sanitation, the ignorance of the population of the most basic hygiene data explain the high degree of the endemicity. Inventory of available means: three types of actions can be considered (Table I-V) (Sharma, 2021)

4. Conclusion

The increasing abundance of parasites in streams and wells in Bafia is a rising sanitary concern. The main protozoan are *Cryptosporidium, Giardia, Cyclospora*, *Strongyloides*, *Ankylostoma* and *Ascari.* Diagnosis and treatment of helminth eggs is a major assessment to be conducted for safe use, reuse of water and wastewater. It is necessary for researchers and scientists to understand protozoa and helminths and develop necessary tools to detect and eradicate them. Proper diagnostic and treatment technologies should be adopted in water reuse schemes to stop the spread of protozoa and helminth eggs in the environment.

Compliance with ethical standards

Disclosure of conflict of interest

No conflict of interest to be disclosed.

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