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Research Article

PREVALENCE AND RISK FACTORS OF PARASITIC PROTOZOAL INFECTIONS IN SCHOOL CHILDREN IN THE KWABRE EAST DISTRICT OF ASHANTI REGION, GHANA

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ABSTRACT
The study aimed to describe the prevalence and distribution of protozoal infections and their predisposing risk factors among pupils in the Kwabre East District, Ghana. Intestinal protozoal infections are a common occurrence among school children, especially in tropical climates and underdeveloped societies owing to poor hygiene and sanitation, overpopulation, illiteracy and low living standards. Fresh stool samples from 884 pupils—one sample per person—representing 48 schools were screened for intestinal protozoa using iodine and saline mounts and the formol-ether concentration technique after questionnaire administration. An isolation rate of 55.7% (492/884) of which 59.6%, 24.8% and 15.7% were *Giardia lamblia, Entamoeba coli* and *Entamoeba hystolytica/dispar* respectively, was obtained. *G. lamblia* infection intensity was highest throughout all the circuits within the district. Parents with low education, buying food from vendors, age, poor hand washing practices after visiting the lavatory, drinking borehole/well water, using pit latrines and not taking anti-protozoal medications were factors that predisposed pupils to intestinal protozoal infections. There was a substantial prevalence of *G. lamblia, E coli* and *E. hystolytica* infections among the Kwabre East District pupils owing to their ages, toilets used, hand washing practices, feeding practices, parents' educational status and sources of drinking water.

Keywords: *G. lamblia, E. coli, E. Hystolytica, Kwabre East District, Protozoal Infections, Epidemiology*

INTRODUCTION
Protozoa are estimated to affect 3.5 billion people worldwide, majority of who are children. Protozoa are intestinal parasites that affect the health and nutritional status of the host. Consequently, infested persons, especially children, are in most cases malnourished (Phiri *et al., 2000;* Ayalew *et al.,* 2011). Besides nutritional status, cognitive behaviour, verbal ability, mental function and physical development are affected by parasitic protozoal infections (Ayalew *et al.,* 2011; Hadidjaja *et al.,* 1998). The severity of these infections are dependent on the type of infecting protozoal species, the nature of the interaction between the parasite and host, the nutritional and immunological status of the host, concurrent infections and the intensity and course of the infection (World Health Organization expert committee, 1987).

Poverty, poor environmental and personal hygiene, overpopulated and underdeveloped societies with poor living conditions, unclean drinking water sources and poor drainage, sewage and waste management systems are factors predisposing persons to intestinal protozoal infections. Hence, protozoal infections are of high prevalence in developing countries (Ayalew *et al.,* 2011; Fabiana and Carolina, 2002; Heidari and Rokni, 2003) , albeit the types and prevalence of these infections differ from country to country and region to region due to differences in social, geographical and environmental factors (Ayalew *et al.,* 2011). *Entamoeba hystolytica/dispar* and *Giardia lamblia* are estimated to infect 50 million and 3 million people respectively worldwide. The prevalence of protozoal infections in Ghana ranges between 0.1 to 89%, depending on the geographical location (Walana *et al.,* 2014).

It is generally agreed by most studies that protozoal infections are common and easily spread among children than adults, owing to their playful and interactive nature and reckless eating habits. This is especially so among male and school-going children below age 8 (Walana *et al.,* 2014). Subsequently, epidemiological studies in several communities have shown the presence of giardosis and other protozoal

out breaks among day-care and school children (Fabiana L and Carolina, 2002; Walana *et al.*, 2014; Maraghi, 1997) . For these reasons, school children are best used for protozoal prevalence studies. The prevalence, intensity and distribution of *G. lamblia, E. hystolytica/dispar* and *Entamoeba coli* were studied among school children in the Kwabre District of the Ashanti Region, Ghana and are herein described.

MATERIALS AND METHODS

Study Area and Sampling

The study was conducted in the Kwabre East District, located within latitudes 60 44' North and longitudes 10 33' to 10 44' West between May 2012 and April 2013. The District has a total land area of 246.8 square kilometres, forming about 1.01% of the total land area of Ashanti Region**.** The District capital, Mamponteng, is approximately 14.5 kilometres from Kumasi, the regional capital, to the north east. Manufacturing and sale of traditional textiles, woodcrafts and artefacts are the major economic activities of the District. The Kwabre East District is divided into 6 circuits by the Ghana Education Service: Aboaso, Ahwiaa, Adanwomase, Antoa, Mamponteng and Ntonso circuits.

The study was conducted in 48 out of 116 nursery and kindergarten schools in the Kwabre East District with male and female pupils below age 6. Most (i.e. 68) of the schools did not cooperate or they provided wrong samples and these were left out. The 48 schools had a total population of 960 registered pupils. A maximum of 20 pupils were selected from each school using systematic sampling. A sample size of 884 pupils was obtained and used for the study.

Questionnaire Administration

Elaborate and validated questionnaires were distributed to the pupils to be sent home to be completed by their parents and guardians. The pupils were oriented to the meaning and purpose of the study and taught how to answer the questions. The pupils, and where necessary the parents, were interviewed in the local language, *Twi*, to complement or clarify the information provided in the questionnaires. Socio-demographic data about the pupils and their parents, their hygienic practices in the household and anti-protozoal medications taken within the last six months were addressed by the questionnaires. Questionnaire administration was done with the help of a field assistant and the interviewing of the pupils and parents were conducted with the aid of the teachers and matrons when the parents came for their wards after close of school.

Sample Collection

Plastic stool containers were provided to the pupils to collect about 2 g of their fresh stool within 24 hours. Stool samples were transported to the laboratory within 30-60 minutes of collection and examined. Samples that could not be examined immediately were stored at 4^{o}C. Iodine and saline mounts (direct technique) and the formol-ether concentration techniques were used to identify the presence and number of *Giardia lamblia, Entamoeba hystolytica* and *Entamoeba coli* motile trophozoites, larvae, cysts and eggs as already described (Ayeh-Kumi *et al.,* 2009; Abu-Madi *et al.,* 2011).

Data Analysis

Microsoft Excel® was used to store and analyse data. The sex, age, family composition, parents educational status, hygienic conditions and practices and the sanitary condition of day-care centers were tabulated and manually analysed to determine their relationship with the intensity, distribution, prevalence and types of protozoal parasitic infections. The number of protozoa cysts, motile trophozoites, larvae and eggs per faecal sample was used to define the intensity of infection as few (1-10), moderate (11-20) and high (21-40).

Ethical Consideration

Informed consents were obtained from the heads of the schools, parents and their class teachers. Ethical clearance for the study was obtained from the School of Medical Sciences, Kwame Nkrumah University of Science and Technology (KNUST).

RESULTS AND DISCUSSION

Results

Study Compliance and Isolation/Infection Rates of Parasites

From a total of 960 children registered from the 48 schools, 884 children participated in the study. Reasons for non-compliance were due to misconceptions about the study, misplaced stool containers and the provision of wrong specimens (urine instead of stool). Some were absent on the day of collection whiles others withdrew without reason. The isolation rate of the parasites was 492 (55.7%) out of 884 collected samples.

Prevalence of Intestinal Parasites

G. lamblia infection was seen in 293 (60%) stool samples representing 293 pupils, *E. coli* was seen in 122 (25%) stool samples representing 122 pupils while *E. Hystolytica/dispar* was seen in 77 (15%) stool samples representing 77 pupils (Table 1). *G. lamblia* was highly recorded at Antoa (58/157), followed by Ntonso (56/112), Ahwiaa (53/139), Adanwomase (48/118), Aboaso (41/215) and Mamponteng (37/143). Ntonso had the highest E. *coli* infection rate (26/112), with Antoa (23/157), Adanwomase (22/118), Aboaso (21/157), Ahwiaa (19/139) and Mamponteng (11/143) following in a descending order. *E. hystolytica* infection was highest in Ahwiaa (20/139), followed by Aboaso (17/215), Adanwomase (16/118), Mamponteng (10/143), Antoa (9/157) and Ntonso (5/112). The overall parasite infection rates were 77.7% (Ntonso), 72.9% (Adanwomase), 66.2% (Ahwiaa), 57.3% (Antoa), 40.6% (Mamponteng) and 36.7% (Aboaso) respectively (Table 1).

Table 1: General Prevalence of Intestinal Parasites Among Pre-School Children within the Kwabre-East District

Species	**Circuits (sample sizes, n)**						**Total infected (n=492) (%)**
	Adanwomase (*n*=118) (%)	**Aboaso (*n*=215) (%)**	**Antoa (*n*=157) (%)**	**Ahwiaa (*n=139*) (%)**	**Ntonso (*n=112*) (%)**	**Mamponten (*n=143*) (%)**	
G. lamblia (%)	48 (40.7)	41 (19.1)	58 (36.9)	53 (38.1)	56 (50)	37 (25.9)	293 (59.6)
E. coli (%)	22 (18.6)	21 (9.8)	23 (14.6)	19 (13.7)	26 (23.2)	11 (7.7)	122 (24.8)
E. hystolytica (%)	16 (13.6)	17 (7.9)	9 (5.7)	20 (14.4)	5 (4.5)	10 (7)	77 (15.7)
Total infected (%)	86 (72.9)	79 (36.7)	90 (57.3)	92 (66.2)	87 (77.7)	58 (40.6)	492 (100)

Intensity of Intestinal Protozoan Infection per District/Circuit

Ntonso circuit had the highest parasite intensity followed by Antoa, Adanwomase, Ahwiaa, Aboaso and Mamponteng circuits respectively (Table 2). In terms of the moderate range (11-20), Aboaso circuit recorded the highest intensity, followed by Ntonso, Ahwiaa, Adanwomase, Antoa and Mamponteng circuits. Lowest parasite intensity (1-10) was highest in Ahwiaa circuit followed by Adanwomase, Antoa, Aboaso, Mamponteng and Ntonso circuits (Table 2).

Table 2: Intensity of Intestinal Protozoan Infection per Circuit Among Pre-School Children within the Kwabre-East District

Circuit/Parasite	Intensity			
	Few (1-10)	**Moderate (11-20)**	**High (21-40)**	**Total**
ABOASO				
G. *Lamblia*	12	14	15	41
E. *coli*	2	17	2	21
E. Hystolytica	8	9	0	17
ANTOA				
G. *Lamblia*	19	9	30	58
E. *coli*	10	9	4	23
E. Hystolytica	1	8	0	9
AHWIAA				
G. *Lamblia*	29	14	10	53
E. *coli*	6	9	4	19
E. Hystolytica	10	6	4	20
ADANWOMASE				
G. *Lamblia*	20	12	16	48
E. *coli*	9	10	3	22
E. Hystolytica	11	5	0	16
NTONSO				
G. *Lamblia*	8	19	29	56
E. *coli*	9	11	6	26
E. Hystolytica	0	4	1	5
MAMPONTENG				
G. *Lamblia*	17	15	5	37
E. *coli*	4	2	5	11
E. Hystolytica	0	5	5	10

Risk Factors for Parasitic Protozoal Infections

The infection rate was highest among pupils of ages 4 to 6 years (275/500), compared to those of ages 1 to 3 years (217/384) (Table 3). Pupils whose parents/guardians had primary education recorded the highest infection (155/328 pupils) rate, followed by illiterate (124/264), secondary (112/174) and tertiary (44/118) education parents (Table 3). Prevalence of parasitic infection was high in children who buy food on the street sometimes (266/460) and in pupils who washed their hands with soap after visiting the toilet

(237/522) though more pupils (285/522) who were not infected practised hand washing with soap after visiting the toilet. Ironically, Infection rate was high in pupils who drank borehole or well (204/356), pipe-borne water (184/356), filtered water (54/356) and stream or river respectively (50/356). A higher infection rate was recorded among pupils who use pit latrines (162/256), Kumasi ventilated pit latrine (KVIP) (130/243) and water closet respectively (106/208). Pupils who had not taken anti-parasitic/protozoal medications in the last three months recorded the highest infection (358/482).

Table 3: Risk Factors Associated with Parasitic Protozoal Infections

Risk Factors	Infected (%)	Non-Infected (%)
AGE (years)		
1-3 (n=384)	217 (56.5)	167 (43.5)
4-6 (n=500)	275 (55)	225 (45)
PARENTS' EDUCATIONAL LEVEL		
Illiterate (n=264)	124 (47)	140 (53)
Primary (n=328)	155 (47.3)	173 (52.7)
Secondary (n=174)	112 (64.4)	62 (35.6)
Tertiary (n=118)	44 (37.3)	74 (62.7)
PUPILS EAT FOOD FROM STREET VENDORS		
Yes (n=185)	97 (52.4)	88 (47.6)
No (n=239)	101 (42.3)	138 (57.7)
Sometimes (n=460)	266 (57.8)	194 (42.2)
WASH HANDS WITH SOAP AFTER VISITING TOILET		
Yes (n=522)	237 (45.4)	285 (54.6)
No (n=138)	82 (59.4)	56 (40.6)
Sometimes (n=224)	130 (58)	94 (42)
DRINKING-WATER SOURCE		
Pipe Borne (n=350)	184 (52.6)	166 (47.4)
Stream/River (n=67)	50 (74.6)	17 (25.4)
Borehole/Well (n=356)	204 (57.3)	152 (42.7)
Filtered water (n=111)	54 (48.6)	57 (51.4)
TYPE OF TOILET USED		
Pit Latrine (n=256)	162 (63.3)	94 (36.7)
Water Closet (n=208)	106 (51)	102 (49)
K.V.I.P. (n=243)	130 (53.5)	113 (46.5)
Others (n=177)	94 (53.1)	83 (46.9)
TAKEN ANTI-PARASITIC DRUGS WITHIN LAST 6 MONTHS?		
Yes (n=402)	141 (35.1)	261 (64.9)
No (n=482)	358 (74.3)	124 (25.7)

Discussion

The study was conducted among 960 registered school children in 48 schools in the Kwabre East District, out of which 884 pupils complied (92.1%). An isolation rate of 55.7% (492/884 collected stool samples) was obtained for all protozoal parasites tested for viz., *G. lamblia* (59.6%), *E. coli* (24.8%) *and E. hystolytica/dispar* (15.7%). The prevalence rate obtained in this study (Table 1) exceeds that obtained in studies conducted in Ghana by Walana and peers (2014) and Ayeh-Kumi *et al.,* (2009), which had prevalence rates of 42.9% (n=2400) and 21.6% (n=204) respectively. However, a study in Ethiopia by Ayalew *et al.,* (2011) among 704 school children far exceeded this study in protozoal infection rates (79.8%). The lower prevalence obtained by Ayeh-Kumi *et al.,* (2009) and Walana *et al.,* (2014) is not surprising as these studies were undertaken in urban cities (Accra and Kumasi) compared to the rural and peri-urban communities of the Kwabre East district, where hygiene, sanitation and protozoal awareness is relatively low. Also, Ayeh-Kumi *et al.*'s study was conducted among adults who are more conscious of their hygiene that preschool children who have a proclivity to playing in the soil.

Furthermore, the infection rates obtained for *G. lamblia, E. coli and E. hystolytica* were higher than that reported by Walana *et al.,* (2014) and Ayeh-Kumi *et al.,* (2009), whose prevalence for these species were 12.2%, 10.3% and 0.2% and 2.0% and 2.0% respectively (*E. coli* was not included in Ayeh-Kumi *et al*.'s study). As concluded by Walana *et al.,* (2014), the rural environment of the Kwabre East District with its attendant sanitation problems, representative of rural communities in tropical environments, is possibly responsible for the higher prevalence rate obtained in this area compared to that obtained in the above mentioned Ghanaian studies.

However, there is a similarity in pattern between this study and other studies in terms of the prevalence rates of *G. lamblia, E. coli and E. hystolytica/dispar* in that they are almost always in a descending order, with *G. lamblia* always being the most prevalent, followed by *E. coli* and *E. hystolytica/dispar* (Ayalew *et al.,* 2011; Walana *et al.,* 2014; Ayeh-Kumi *et al.,* 2009; Ahsun-ul-Wadood *et al.,* 2005). The intensity of *G. lamblia* infections in each circuit, as compared to that of the other species, augments this conclusion (Table 2), demonstrating the widespread distribution of *G. lamblia* among school children in tropical and temperate environments. The epizoonotic nature of *G. lamblia* infections suggests the possible interaction between animals and the pupils although pupil interaction with animals were not addressed in the questionnaires. However, the proclivity of pre-school children to play in soils in a rural and peri-urban environment where animals roam and defecate in free range is enough to suggest that these pupils might have picked up the *G. lambia* from animal faecal-contaminated soils. Given the poor fat malabsorption and dysentery, diarrhoea and liver abscess associated with persistent *G. lamblia* and *E. hystolytica/dispar* infections respectively, (Walana *et al.,* 2014) a better intervention should be put in place to reduce the prevalence rate and safeguard the pupils health.

The findings of this study corroborate that of other findings in terms of the risk factors predisposing children to protozoal infections. Pupils between ages 4-6 had higher infections (275/500) than those of ages 1-3 (217/384) albeit the former had a higher percentage. This, as explained elsewhere (Walana *et al.,* 2014), is as a result of the explorative nature of this age group in playing with soils and peers that might be contaminated. The educational level of parents affected the rate of infection with protozoa parasites (Table 3) in that, highly-educated parents were more informed on the importance of hygiene. Moreover, highly educated parents also enjoyed a higher standard of living, which reduced the exposure of their wards to squalid environments (Ayalew *et al.,* 2011). Due to the poor sanitation and waste management systems present in rural communities, children are easily exposed to faeces-contaminated environments and drinking water. Hence, the higher infection rate observed among pupils who used pit latrines and borehole/well water (Table 3). Especially as the borehole/well water can be contaminated with flies, insects and reptiles that patronise the faeces in the pit latrines (Ayeh-Kumi *et al.,* 2009). Therefore, it is no surprise that most of the non-infected pupils were those who washed their hands with soap after visiting the toilet (Table 3). Also, the relatively higher number of pupils who were infected with protozoa in spite of washing their hands with soap could suggest that these pupils were infected from other sources besides their fingers or that they did not wash their hands thoroughly after visiting the toilet. The

importance of taking anti-protozoal medications is seen in Table 3, with most pupils who took these medications having fewer infections. The role of food vendors in transmitting protozoal parasites, as already discussed (Ayeh-Kumi *et al.,* 2009) is a major risk factor (Table 3) for the pupils who easily buy food from these vendors.

Conclusion

There is a substantial prevalence of *G. lamblia, E. coli* and *E. hystolytica* infections among pupils in the Kwabre East District of the Ashanti Region, Ghana. Parents' educational status, hygienic practices, source of drinking water, toilet facilities used, history of anti-protozoal medications and age are factors predisposing the pupils to intestinal protozoal infections.

Authors' Contributions

ABC and OSJ conceived research design, OSJ drafted paper. ABC undertook laboratory experiments. Both authors read and approved the manuscript for submission.

Competing Interests

The authors declare that they have no competing interests.

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