TMJ
Original research
Published by OJS
Doi: 10.4314/tmj.v32i2.429.g265
OPEN ACCESS JOURNAL

Prevalence of Intestinal Protozoa Infections and Associated Risk Factors among Preschool Aged Children in Mkuranga District, Tanzania: A Community Based Cross-Sectional Study

Ummul-Khair Mustafa[1], Abdallah Zacharia[2]*, Vivian Mushi[2]

[1]Department of Biological Science, Faculty of Science, Dar es Salaam University College of Education, Dar es Salaam, Tanzania
[2]Department of Parasitology and Medical Entomology, Muhimbili University of Health and Allied Sciences, Dar es Salaam, Tanzania

***Corresponding author:**
Abdallah Zacharia
Muhimbili University of Health and Allied Sciences
P. O. Box 65011
Dar es Salaam, Tanzania
Email: naayz@ymail.com

TMJ
Original research
Published by OJS
Doi: 10.4314/tmj.v32i2.429.g265
OPEN ACCESS JOURNAL

Abstract

Background
Intestinal protozoa infections are among the public health concern in children and can lead to considerable morbidities such as growth retardation, anemia, nutrition deficiency, physical weakness and mental health problems. In Tanzania, there is a paucity of data on intestinal protozoa infections among preschool aged children (PSAC).

Broad objective
This study aimed to determine the prevalence of intestinal protozoa infections among PSAC and associated risk factors in Mkuranga district, Tanzania.

Methodology
A community based cross-sectional study was conducted between April and June 2019 to determine the prevalence of intestinal protozoa. Stool samples were collected from PSAC and processed using formal-ether concentration method. Risk factors were identified using questionnaire and checklist. Data were entered and analysed by using Statistical Package for the Social Sciences software version 20. Univariate and multivariate logistic regressions were performed to determine association between prevalence of intestinal protozoa and socio-economic, water source, hygiene and sanitation variables.

Results
A total of 20 (3.8%) children were infected with intestinal protozoa (*Entamoeba coli* or *Giardia lamblia*). We found the increased risk of intestinal protozoa infections with increased age of children (AOR = 1.07, p = 0.045). Also, we observed a reduced odds of intestinal protozoa infections among PSAC living in low populated households (AOR 1-5/11+ = 0.14, p = 0.016 and AOR 6-10/11+= 0.10, p = 0.008), using non-spring water (AOR protected well/spring = 0.011, p = 0.023), (AOR bore hole/spring/spring = 0.008, p = 0.006), (AOR tap away of home/spring = 0.013, p = 0.014) and (AOR tape at home/spring = 0.006, p = 0.011)) and raised by young and middle-aged parents/guardian (AOR young/older = 0.20, p = 0.042 and AOR middle-aged/older = 0.18, p = 0.047).

Conclusion
There was a low prevalence of intestinal protozoa infection among PSAC in the two communities studied. Despite low prevalence of intestinal protozoa, the transmission is present hence the need to initiate control measures. Health education should be provided to parents/guardians on protecting water sources, treatment of drinking water and supervision of latrine usage by PSAC and introduction of hand washing facilities are crucial.

Key words: *Intestinal protozoa, Preschool aged children, Mkuranga and Tanzania.*

Published by OJS
Doi: 10.4314/tmj.v32i2.429.g265

Introduction

Intestinal protozoa infections are endemic in tropical and subtropical regions with limited access to clean water, poor sanitation, and hygiene (1). The intestinal protozoa infections are more prevalent to preschool aged children (PSAC) and school aged children where they contribute to significant morbidity and mortality (2,3). Children are more susceptible to intestinal protozoa infections because of less developed immune system and their behaviors such as playing in fecal contaminated soil, eating with unwashed hands, drinking unsafe water, eating contaminated food and unpeeled fruits(4). Intestinal protozoa infections in children can cause growth retardation, anemia, nutrition deficiency and physical weakness (3,5). The intestinal protozoa infections in children can be asymptomatic or with mild symptoms; protozoa such as *Entamoeba histolytica, Giardia* and *Cryptosporidium parvum* can cause severe dysentery and diarrhea (6).

There is a significant burden of intestinal protozoa globally. Data collected in 2010 showed that 64, 104, and 184 million people suffered from *Cryptosporidium* species, *Entamoeba histolytica*, and *Giardia* species, respectively leading to more than 30,000 deaths (7). Additionally, 0.43 and 4.0% of global deaths in children less than five years were due to amoebiasis and cryptosporidiosis, respectively (8). Moreover, amoebic colitis account for 9% of all under five children deaths in low-income counties (9). In Sub-Saharan Africa the prevalence of intestinal protozoa in children has been reported to range from 16%-90% in different geographical settings with different distribution of protozoan species. Evidence shows that PSAC are capable of maintaining transmission in the community (10).

In Tanzania, little is known about the prevalence of intestinal protozoa in PSAC and their associated risk factors. A study conducted among school-aged children in Pemba (Tanzania) showed a high prevalence of intestinal protozoa infections (74.7%). The high prevalence of intestinal protozoa was due to negligence because the community considers the infections of less significance (3). Another study conducted among children under five years who presented with diarrhoea in three hospitals in Dar es Salaam revealed an overall prevalence of intestinal protozoa infections (*Giardia lamblia, Entamoeba histolytica* and *Cryptosporidium spp*) to be 41% (11). Therefore, this study was conducted to determine the prevalence of intestinal protozoa infections and associated risk factors among PSAC in rural and suburban communities of Mkuranga district, Tanzania.

Methods

Description of the study area

Mkuranga is one of the six districts in Pwani region.; the district boarders with Dar es Salaam region, Indian Ocean, Rufiji district and Kisarawe district to the north, east, south and west respectively. According to the population and housing census of 2012, the district has a total population of 222,921 of which males were 108,024 and females were 114,897(12). Mkuranga was selected because of high prevalence (32.7%) of underfive diarrhea and inadequate water, sanitation and hygiene conditions among many households in the district (13). Additionally, Mkuranga district has both rural and sub-urban settings providing an ideal study site based on our objective. Mwarusembe and Kazole villages were selected from Mwarusembe and Vikindu wards, respectively. As per 2012 census, Mwarusembe and Vikindu wards had a population size of 11,706 and 34,762 with an average of 4.2 and 4.5 members per household, respectively (12). Mwarusembe village is a rural setting located 70.5 km south of Dar es Salaam, the country's largest economy city. Kazole village is sub-urban setting located 27.2 km south of Dar es Salaam (14).

Study design and population

A community based cross-sectional study involving quantitative methods was carried out from April to June 2019. This study was part of the large study that investigated intestinal parasitic infections among PSAC in Mkuranga District. Study participants were PSAC for the prevalence of intestinal protozoa and parents/guardian for assessing risk factors associated with transmission of intestinal protozoa. PSAC included children not enrolled in primary school and aged between 12 and 59 months (15).

Sample size and sampling procedure

The sample size was calculated from a formula which is given by Creswell(16). The estimated sample size for this study was 612 PSAC from 612 households. Each PSAC was accompanied by his/her parent/guardian. Participants were obtained through four-stage cluster sampling technique as indicated in figure 1. We obtained the number of households with eligible child/children in each hamlet from hamlet leaders. The number of households contributed by each hamlet to the estimated sample size was calculated by dividing the number of houses with eligible child/children in a particular hamlet by the total number of houses with eligible child/children in the two hamlets times the estimated sample size (612 households). We identified the center of the hamlet as a starting point. The researcher spun

a bottle to determine the direction. We selected the household closest to the center and pointed by the bottle as the first household. We took every next nearest house in the determined direction until we achieved the estimated sample. In a household with more than one eligible child, only one was selected using the lottery method.

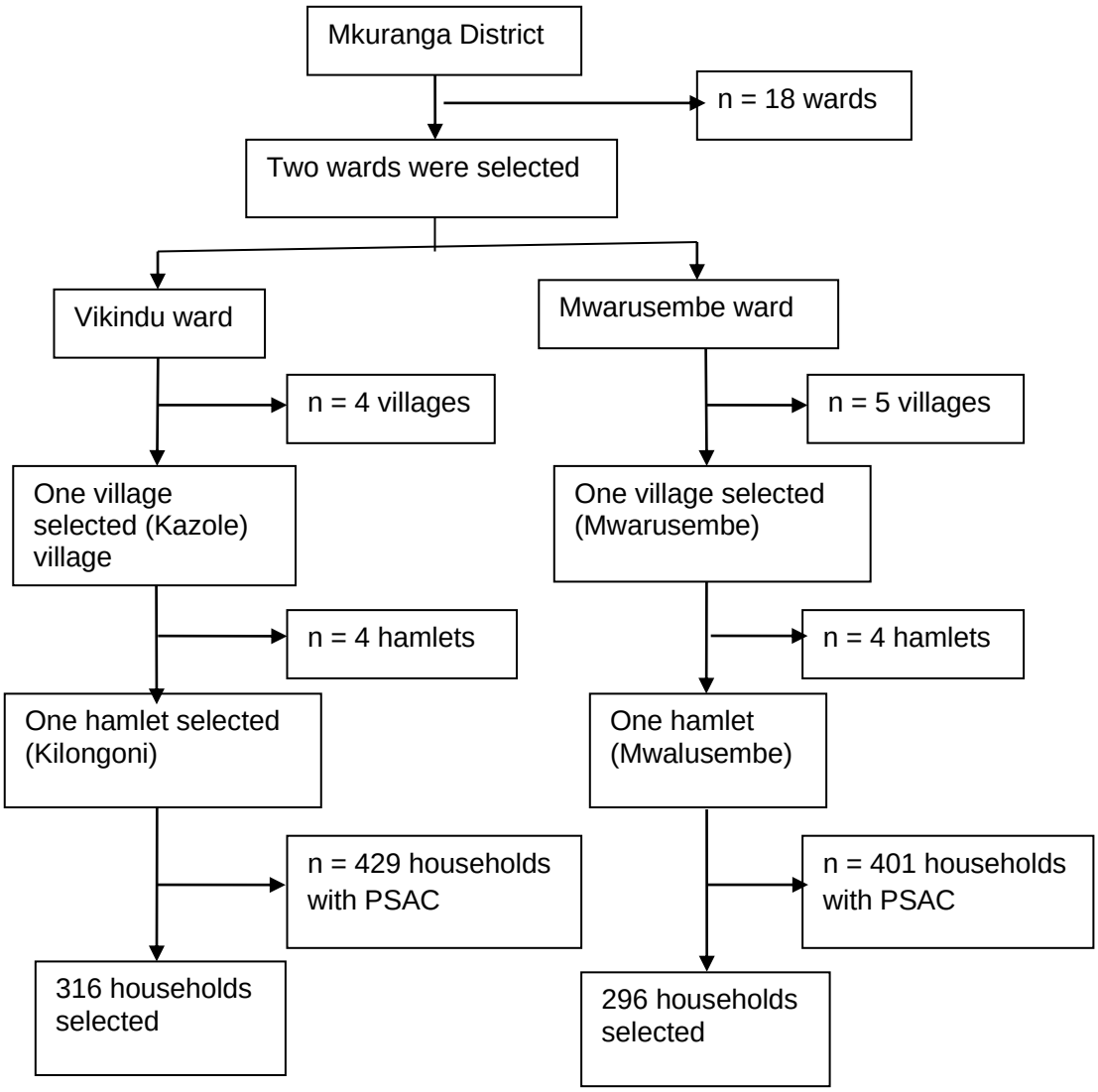

Figure 1: Flow chart showing sampling stages

Sample collection and laboratory analysis

To determine the prevalence of intestinal protozoa; stool samples were collected, processed and examined for presence or absence of intestinal protozoa oocysts, cysts and/or trophozoites. Consented parents were instructed to let the child defecate on the provided

clean piece of paper, and use an applicator stick to put a portion of stool into a given clean, dry and wide mouthed stool container. Collected stool sample was stored in a safe place (out of reach of children) to wait for research assistance to pick. We instructed parents/guardians on the importance of proper fecal disposal and handwashing methods after the sample collection. Collected samples were transported to the Parasitology laboratory at the Muhimbili University of Health and Allied Sciences and examined within 24 hours (17). In situations where samples could not be examined in the day of collection, they were preserved in 10% formalin (one volume of stool sample in three volumes of formalin) (18). A single stool sample was collected per each PSAC. Stool specimens were processed using formal-ether concentration techniques as described in the World Health Organization (WHO) bench aids for the diagnosis of parasitic diseases (19). Briefly, about 1 g of feces was mixed with 10 mL of fixative (formalin 5–10%) and left for at least 30 minutes. The suspension was strained in a 15ml centrifuge tube using double-layer gauze and centrifuged at 500 *g* for 10 minutes. We removed the supernatant and added 7 ml of saline to the sediment. We sealed the tube with a stopper and mixed it. Then we added 3 ml of ethyl acetate and shaken it vigorously for 30 seconds. We left the centrifuge tube for 15-30 seconds, carefully opened, and centrifuged at 500 *g* for 3 minutes. We detached the plug of debris from the tube wall with an applicator stick, and then poured it off and the sediment mixed with the remaining liquid. We placed a drop of sediment on a slide and covered it with a coverslip. Also, on the same microscope slide, we prepared a Lugol's Iodine stain smear. Examination for the presence of intestinal protozoa oocysts, cysts and/or trophozoites was done using Olympus CX 31 microscope.

Questionnaire survey

A questionnaire with structured questions supported with observation checklist was used to collect demographic characteristics of participants and possible risk factors associated with protozoa transmission such as water source, hygiene and sanitation conditions. All sections of questionnaire were administered to interviewees (parents/guardians) by trained research assistants.

Inclusion and exclusion criteria

PSAC living in the study area for the past six months and whose parents/guardians signed the written consent were eligible to participate. Those who received anti-intestinal protozoa drugs such as metronidazole within one month prior to data collection, children having

diarrhea at the time of stool collection, children whose parents/guardians refused to sign a written consent and children aged 5 years but attending primary schools were excluded from the study.

Data analysis

Data collected were cleaned, coded and entered into Statistical Package for Social Sciences version 23 for analysis. The categorical data were summarized using frequency and proportion while mean and standard deviation used for continuous data. Univariate logistic regression was performed to determine association between prevalence of intestinal protozoa and socio-economic and environmental variables such as education of parents/guardians, economic status (poverty), water, sanitation, and hygiene conditions. Variables shown to have statistical significant association during univariate logistic regression analysis were subjected to multivariate logistic regression test. P-value less than 0.05 was considered statistically significant.

Ethical considerations

Ethical clearance was sought from the Muhimbili University of Health and Allied Sciences Senate Research and Publications Committee. Permission was obtained at all administrative units involved in this study, from the regional level to the hamlet level where this study was conducted. A written consent was requested from the parents/guardians of each participant. Infected children were referred to the nearest dispensaries immediately after laboratory results were out. A summary of results/notification were reported to the medical district officer for records.

Results

A total of 525 out of 612 heads of households from the two selected villages consented to participate in the study. In each consented household, we selected one PSAC and one parent/guardian. The children's mean age was 36.7 (±14.3) months ranged from 12 to 59 months. Female children were more than half (52.4%) of all children participants. The mean age of parents/guardians was 30.8 (±9.1) years and ranged between 16 to 70 years. Young parents/guardians (< 36 years old) contributed a high number (75%) of parents/guardians compared to the other two groups. Occupations of the interviewed parents/guardians descend in the following order: unemployed, self-employed, peasant, and private or public

employee. Sixty-three percent of all parents/guardians attained primary school education level, and 19.6% lacked any formal education (Table 1).

The prevalence of intestinal protozoa infection was 3.8% in which two species were identified. The two species were *Entamoeba coli* (2.3%) and *Giardia lamblia* (1.5%). Females were more infected than males with prevalence of 4.7% and 2.8% respectively. However, none of the children was found co-infected with the two intestinal protozoa species. The prevalence of intestinal protozoa was high in children raised by older parents/guardians (56 – 75 years), employed, with secondary school education level, and high economic status. The high prevalence was also found in children living in households with high number of household members (11+), households without latrines and households without hand washing facilities. One out of 2 PSAC whose parents/guardians reported that they use spring water were infected with *Giardia lamblia* (Table 1). All infected PSAC were from households with easy access of water (regardless of the quality), do not treat drinking water and without hand washing facilities after toilet.

Table 1: Social-demographic characteristics of study participants and assessed risk factors

Characteristic	Categories	No of PSAC	No of Infected PSAC (%)
Sex of PSAC	Female	275	13 (4.7)
	Male	250	7 (2.8)
Study settings	Rural	254	9 (3.5)
	Suburban	271	11 (4.1)
Age groups of parents/guardians	Young (< 36 years)	392	13 (3.3)
	Middle-aged (36-55 years)	124	5 (4.0)
	Older (56-75 years)	9	2 (22.2)
Occupation of parents/guardians	Employed	29	2 (6.9)
	Self- employee	158	6 (3.8)
	Peasant	96	3 (3.1)
	Unemployed	242	9 (3.7)
Economic status of parents	Low	195	5 (2.6)
	High	330	15 (4.5)
Education level of parents/guardians	Never attended school	103	3 (2.9)
	Primary education	332	12 (3.6)
	Secondary education	67	4 (6.0)
	Tertiary education	23	1 (4.3)
Households' population	11+	13	3 (23.1)
	6 – 10	194	5 (2.6)

	1 -5	318	12 (3.8)
Water availability	Difficult	32	0 (0.0)
	Easy	493	20 (4.1)
Source of water for domestic activities	Spring	2	1 (50.0)
	Unprotected well	24	0 (0.0)
	Protected well	27	1 (3.7)
	Tube well or bore hole	207	8 (3.9)
	Tape away of home	224	9 (4.0)
	Tape at home	41	1 (2.4)
Drinking water treatment	No	28	1 (3.6)
	Yes	497	19 (3.8)
Hand washing facility	Present	7	0 (0.0)
	Absent	518	20 (3.9)
Nails of PSAC	Untrimmed	87	2 (2.3)
	Trimmed	458	18 (3.9)
Latrine availability	Not available	14	2 (14.2)
	Available	493	18 (3.7)
Latrine use by PSAC	Use latrine	245	17 (6.9)
	Not use latrine	280	3 (1.1)

Note: PSAC = preschool aged children

Table 2 summarizes the results of univariate and multivariate logistic regression tests. The test indicated statistically significant association between protozoa prevalence and age of children, age group of parents/guardians, households' population and latrine use. When these factors were adjusted for the confounders using multivariate logistic regression, all were significantly associated with intestinal protozoa prevalence except for the latrine use. After analysing children age as a continuous variable, the findings showed that the odds of intestinal protozoa infections among children increase with an increase in age. Also, we found that there is less risk of intestinal protozoa infection among children with young and middle-aged parents/guardians than those with older parents/guardians. Moreover, there is a risk reduction of intestinal protozoa infection among children using tap water (away or at home), tube hole (borehole) water, and water from protected wells compared to those using spring water.

Published by OJS
Doi: 10.4314/tmj.v32i2.429.g265

Table 2: Association between intestinal protozoa infection and risk factors (socioeconomic, behavioral, water, hygiene and sanitations)

Characteristic	Categories	COR (95% CI)	P-value	AOR (95% CI)	P-value
Age of PSAC	NA	1.09 (1.04 – 1.14)	< 0.001	1.07 (1.00 – 1.13)	0.045
Sex of PSAC	Male	1		NA	NA
	Female	1.72 (0.68 – 4.39)	0.254		
Study settings	Rural	1		NA	NA
	Suburban	0.87 (0.35 – 2.13)	0.758		
Age groups of parents or guardians	Older (56-75 years)	1		1	
	Young (< 36 years)	0.30 (0.06 – 1.62)	0.013	0.20 (0.03 – 1.23)	0.042
	Middle-aged (36-55 years)	0.15 (0.03 – 0.74)	0.038	0.18 (0.03 – 1.28)	0.047
Occupation of parents or guardians	House wife	1		NA	NA
	Self-employee	1.02 (0.36 – 2.93)	0.968		
	Peasant	0.84 (0.22 – 3.15)	0.790		
	Public/private employee	1.92 (0.39 – 9.34)	0.420		
Economic status (Classified based on house condition)	Good	1		NA	NA
	Low	0.55 (0.20 – 1.55)	0.258		
Level of education of parents or guardian	Tertiary education	1		NA	NA
	Secondary education	1.40 (0.15 – 13.13)	0.770		

Published by OJS
Doi: 10.4314/tmj.v32i2.429.g265

	Primary education	0.83 (0.10 – 6.64)	0.857		
	Never attended school	0.66 (0.07 – 6.65)	0.724		
Households' population	11+	1		1	
	6 – 10	0.09 (0.02 – 0.42)	0.002	0.10 (0.02 – 0.54)	0.008
	1 – 5	0.13 (0.03 – 0.54)	0.005	0.14 (0.03 – 0.69)	0.016
Source of water for domestic activities such as drinking and washing.	Spring	1		1	
	Unprotected well	0.000	0.998	0.000	0.998
	Protected well	0.038 (0.001 – 1.2)	0.062	0.011 (0.000 – 0.5)	0.023
	Tube well/bore hole	0.040 (0.002 – 0.7)	0.028	0.008 (0.000 – 0.25)	0.006
	Tape water away of home	0.042 (0.002 – 0.7)	0.029	0.013 (0.000 – 0.4)	0.014
	Tape water at home	0.025 (0.001 – 0.8)	0.034	0.006 (0.000 – 0.3)	0.011
Nails of PSAC	Trimmed	1		NA	NA
	Untrimmed	0. 75 (0.17 – 3.32)	0.707		
Latrine availability	Available	1		NA	NA
	Not available	4.57 (0.95 – 21.92)	0.058		
Latrine use by PSAC	Not use	1		1	
	Use	6.89 (1.99 – 23.79)	0.002	3.05 (0.51 – 18.13)	0.220

Note: COR = crude odd ratio, CI = confidence interval, AOR = adjusted odd ratio, PSAC = preschool aged children and NA = not applicable.

Published by OJS
Doi: 10.4314/tmj.v32i2.429.g265

Discussion

Findings of this study indicated that overall prevalence of intestinal protozoa is low among PSAC in Mkuranga district. The overall intestinal protozoa prevalence in this study is lower compared to the prevalence recorded among under five children with diarrhea in Dar es Salaam, a region located few kilometers from Mkuranga district (11). However, the prevalence reported in Dar es Salaam was based on children already with symptoms of intestinal protozoa infection (diarrhea) unlike asymptomatic children recruited in the present study. The prevalence of non-pathogenic intestinal protozoa (*Entamoeba coli*) was high compared to that of pathogenic intestinal protozoa (*Giardia lamblia*). Similar results were found in a study conducted on school-going children in Zanzibar whereby non-pathogenic intestinal protozoa were predominantly found compared to the pathogenic one (3). The predominance of non-pathogenic intestinal protozoa was most likely because parents or guardians of infected children did not seek treatment as the children were asymptomatic. The presence of non-pathogenic intestinal protozoa is an indicator of poor sanitary and hygienic condition in the studied community (20).

Infection with intestinal protozoa were reported to be associated with several factors such as age and sex of children, age and sex of parents/guardians, education of parents/guardians, economic status of parent/guardians, place of residence, water source for domestic activities, personal hygiene and sanitation conditions (7). In the current, study 13 variables describing the above mentioned risk factors were assessed for their contribution to the prevalence of intestinal protozoa (Table 1 and 2). The risk of intestinal protozoa infection was observed to significantly increase with an increase in the age of children (Table 2). This is similar to the results obtained from studies conducted in Ethiopia whereby intestinal parasitic infections (including protozoa) were found to increase with age of the children (21,22). The reason could be due to the fact that child age had an influence on the health seeking behavior of the parents/guardian. A study conducted in three districts in Tanzania reported that parents/guardians are more likely to seek health care for young children compared to older children (23). PSAC raised by older parents/guardians were significantly more likely to have intestinal protozoa infections than those raised by parents/guardians in other age groups. This supports the hypothesis that older mothers/parents/guardians are less concerned with the health of their children compared to young

mothers/parents/guardians (24). Also, diarrhea is a predominant symptom of most intestinal protozoa infections (25).

However, a study conducted in Tanzania revealed that older parents/guardians perceived diarrhea as a non-serious illness thus do not seek immediate health care incase their children had diarrhea (23). Increase in number of household members was seen to increase the risk of intestinal protozoa infection which is similar to the findings from a study conducted in Ethiopia (26). The higher risk of intestinal protozoa in households with a large number of family members possibly was attributed to the fact that health-seeking behaviors in large-sized families are low compared to small-sized families (24). The decreased in health-seeking behaviors in large-sized families could be due to limited available resources which should be invested in all family members.

In the two study communities, water availability and latrine coverage were very high. A total of six water sources were reported by study participants. Moreover, about half of the PSAC population was observed to utilize latrines during defecation (Table 1). However, spring water sources and latrine utilization were significantly associated with the prevalence of intestinal protozoa (Table 2). The parents/guardians who reported to use spring water had a significantly high prevalence of intestinal protozoa than those reported to use piped water. Majority parents/guardians declared that they didn't treat water for drinking (Table 1). This behavior increases the chance of intestinal protozoa transmission especially if the sources of water are not safe. Similar results were found in a study conducted in Iran (27). We expected that PSAC who utilized latrines for defecation to be less infected with intestinal protozoa than those not using latrines (use alternative facilities such as dippers/pants, defecating on the ground or the children chamber pots). But our findings showed that the PSAC using latrines were significantly affected by intestinal protozoa than those not using the latrine facilities (use alternative facilities such as dippers/pants, defecating on the ground or children's chamber pots). The possible reason for this could be that, PSAC who used latrines were not supervised by their parents/guardians during the process and therefore didn't adhere to the hygienic practices such as proper hand washing after latrine use and anal cleansing. Latrine use and care by themselves (children) was associated with high intestinal parasitic infection (including protozoa) in Ethiopia (21). In addition, more than 90% of latrines in the two studied communities had no hand washing facilities that have water and soap either inside or outside of the latrine. The presence of hand washing facilities surrounding the toilets

influences the behavior of hand washing. Moreover, it is difficult for PSAC to find water and soap for washing their hands if it is not located nearby.

Conclusion and recommendation

The results of this study show that the prevalence of intestinal protozoa among PSAC is low. Despite the low prevalence, it's clear that the transmission is ongoing. The risk factors associated with intestinal protozoa infections among PSAC in the two communities were latrine usage by PSAC and spring water sources. Therefore, health education on the transmission of intestinal protozoa is required among the parents/guardians of PSAC in the two communities. The health education package should include how to assist PSAC during latrine use, water treatment for drinking and introduction of hand washing facility after defection.

Study limitation

The use of the formal ether technique alone may underestimate the prevalence of intestinal parasite infections as it is not good for the diagnosis of coccidian intestinal protozoa.

List of abbreviation

AOR	Adjusted Odd Ratio
COR	Crude Odd Ratio
PSAC	Pre-school aged children
WHO	World Health Organization

Competing interests

The authors declare that they have no competing interests.

Data availability

The data used to support the findings of this study are available from the corresponding author upon request.

Funding

This work was funded by Mwalimu Julius Nyerere Memorial scholarship fund (Bank of Tanzania). The fund covered expenses for sample collection and laboratory analysis. The

Published by OJS
Doi: 10.4314/tmj.v32i2.429.g265

authors declare that no other financial support was secured from other sources for this research work.

Authors' contributions

UM designed the study and collected data, UM and AZ performed laboratory analysis, AZ and VM performed statistical analysis, AZ and VM wrote the manuscript. All authors read and approved the final manuscript.

Acknowledgements

The authors wish to thank Mr. Badi. H. Badi, Mr. Rajab Mnyangema, and Ms Mary Joseph for their assistance in data collection and sample processing. Special thanks go to Ward and village leaders.

Published by OJS
Doi: 10.4314/tmj.v32i2.429.g265

References

1. Ayalew A, Debebe T, Worku A. **Prevalence and risk factors of intestinal parasites among Delgi school children, North Gondar, Ethiopia.** J Parasitol Vector Biol. 2011;3(5):75–81.
2. Hotez PJ, Alvarado M, Basáñez M-G, Bolliger I, Bourne R, Boussinesq M, et al. **The global burden of disease study 2010: Interpretation and implications for the neglected tropical diseases.** PLoS Negl Trop Dis. 2014;8(7):e2865.
3. Speich B, Marti H, Ame SM, Ali SM, Bogoch II, Utzinger J, et al. **Prevalence of intestinal protozoa infection among school-aged children on Pemba island, Tanzania, and effect of single-dose albendazole, nitazoxanide and albendazole-nitazoxanide.** Parasit Vectors. 2013;6(1):3.
4. Sitotaw B, Mekuriaw H, Damtie D. **Prevalence of intestinal parasitic infections and associated risk factors among Jawi primary school children, Jawi town, north-west Ethiopia.** BMC Infect Dis. 2019;19(341):1–10.
5. Tulu B, Taye S, Amsalu E. **Prevalence and its associated risk factors of intestinal parasitic infections among Yadot primary school children of South Eastern Ethiopia: a cross-sectional study.** BMC Res Notes. 2014;7(848):1–7.
6. Galgamuwa L, Iddawela D, Dharmaratne S. **Intestinal protozoa infections, associated risk factors and clinical features among children in a low-income tea plantation community in Sri Lanka.** Int J Community Med Public Heal. 2016;3(9):2452–8.
7. Kirk MD, Pires SM, Black RE, Caipo M, Crump JA, Devleesschauwer B, et al. **World health organization estimates of the global and regional disease burden of 22 foodborne bacterial, protozoal, and viral diseases, 2010: A data synthesis.** PLoS Med. 2015;12(12):1–21.
8. Lozano R, Naghavi M, Foreman K, Lim S, Shibuya K, Aboyans V, et al. **Global and regional mortality from 235 causes of death for 20 age groups in 1990 and 2010: A systematic analysis for the Global Burden of Disease Study 2010.** Lancet. 2012;380(9859):2095–128.

9. Shirley DT, Farr L, Watanabe K, Moonah S. **A review of the global burden, new diagnostics, and current therapeutics for amebiasis.** Open Forum Infect Dis. 2018;5(7):1–9.
10. Walana W, Crowther S, Tay K, Tetteh P, Ziem JB. **Prevalence of intestinal protozoan infestation among primary school children in urban and peri-urban communities in Kumasi, Ghana.** Sci J Public Heal. 2014;2(2):52–7.
11. Ngosso BE, Nkwengulila G, Namkinga LA. **Identification of pathogenic intestinal parasitic protozoa associated with diarrhea among under-fives children in Dar es Salaam, Tanzania.** Int Invent J Med Med Sci. 2015;2(4):49–55.
12. United Republic of Tanzania. **2012 population and housing census population distribution by administrative areas.** National Bureau of Statistics, Ministry of Finance. 2013.
13. Kakulu RK. **Diarrhoea among underfive children and household water treatment and safe storage factors in Mkuranga district.** Muhimbili University of Health and Allied Sciences; 2012.
14. Google. **Google map** [Internet]. 2020 [cited 2020 Jun 28]. Available from: https://www.google.com/maps/dir///@-6.883992,39.1088331,11z
15. Harris JR, Worrell CM, Davis SM, Odero K, Mogeni OD, Deming MS, et al. **Unprogrammed deworming in the Kiberaslum, Nairobi: implications for control of soil-transmitted helminthiases.** PLoS Negl Trop Dis. 2015;9(3):1–11.
16. Creswell JW. **Research Design: Qualitative, Quantitative and Mixed Method Aproaches.** SAGE Publ. 2007;
17. Garcia LS, Arrowood M, Kokoskin G, Paltrdge GP, Pillai DR, Procop GW, et al. **Laboratory diagnosis of parasites from the gastrointestinal tract**. Clin Microbiol Rev. 2018;31(e00025-17).
18. CDC. **DPDx-Laboratory Identification of Parasites of Public Health Concern: stool specimens - specimencollection** [Internet]. 2016 [cited 2021 Jan 10]. Available from: https://www.cdc.gov/dpdx/diagnosticprocedures/stool/specimencoll.html
19. **Bench aids for diagnosis of intestinal parasites.** second edi. Geneve: World Health Organization; 2019.
20. Issa R. **Non-pathogenic protozoa (Review article)**. Int J Pharm Pharm Sci. 2014;6(3):30–10.
21. Gadisa E, Jote K. **Prevalence and factors associated with intestinal parasitic**

infection among under-five children in and around Haro Dumal Town, Bale Zone, Ethiopia. BMC Pediatr. 2019;19(1):1–8.

22. Aleka Y, G/egziabher S, Tamir W, Birhane M, Alemu A.**Prevalence and associated risk factors of intestinal parasitic infection among under five children in University of Gondar hospital, Gondar, Northwest Ethiopia.** Biomed Res Ther. 2015;2(8):347–53.
23. Kanté AM, Gutierrez HR, Larsen AM, Jackson EF, Helleringer S, Exavery A, et al. **Childhood illness prevalence and health seeking behavior patterns in rural Tanzania.** BMC Public Health. 2015;15(1):1–12.
24. Uggla C, Mace R. **Parental investment in child health in sub-Saharan Africa: a cross-national study of health-seeking behaviour.** R Soc Open Sci. 2016;3(150460):1–13.
25. Bauhofer AFL, Cossa-Moiane I, Marques S, Guimarães EL, Munlela B, Anapakala E, et al. **Intestinal protozoan infections among children 0-168 months with diarrhea in Mozambique:** June 2014-january 2018. PLoS Negl Trop Dis. 2020;14(4):1–17.
26. Gizaw Z, Addisu A, Gebrehwot M. **Socioeconomic predictors of intestinal parasitic infections among under-five children in rural Dembiya, Northwest Ethiopia: a community-based cross-sectional study.** Enviromental Heal Insights. 2019;13:1–6.
27. Abbaszadeh Afshar MJ, Barkhori Mehni M, Rezaeian M, Mohebali M, Baigi V, Amiri S, et al. **Prevalence and associated risk factors of human intestinal parasitic infections: a population-based study in the southeast of Kerman province, southeastern Iran.** BMC Infect Dis. 2020;20(1):1–8.