Tanzania Journal of Science 46(2): 548-563, 2020
ISSN 0856-1761, e-ISSN 2507-7961

Current Epidemiological Assessment of *Plasmodium falciparum* and Helminth Co-Infections in Children after a Decade of Implementation of Control Programs in Morogoro Region, Tanzania

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Received 8 May 2020, Revised 25 June 2020, Accepted 27 June 2020, Published June 2020

Abstract

A school based cross-sectional study was conducted from July to November 2018 to assess the burden of asymptomatic *Plasmodium falciparum*, *Schistosoma* and soil transmitted helminth (STH) infections in Mvomero. A total of 374 children (age range = 5–16 years, mean age = 11.3 years) were recruited from five primary schools. Prevalence of asymptomatic *P. falciparum* infections were 29.9%, *S. haematobium* 49.7%, hookworm 20.3%, *Ascaris lumbricoides* 12.6%, *Taenia saginata* 0.5% and *S. mansoni* 0.3%. Malaria parasite density increased with increasing children age (r = 0.99). Only 6.5% (12/186) of *S. haematobium* infected children were presented with heavy infections, whereas all STH-positive children harboured light infections. The overall *P. falciparum-helminths* co-infection rate was 11%. *Schistosoma haematobium* and hookworm infections positively influenced *P. falciparum* parasitaemia (R^2 = 0.55 and 0.73, respectively). Being between 11 and 13 years of age, father being a farmer, poor housing, not sleeping under insecticide treated net, working in rice and sugarcane fields were the major factors associated with asymptomatic *P. falciparum*-helminth co-infections (all $p < 0.05$). Prevalence of both asymptomatic *P. falciparum* infections and *P. falciparum*-helminths-co-infections has decreased by over 40%. However prevalence of *S. haematobium* and hookworm infections is alarmingly high, calling for community based-integrative control measures incorporating strategies to combat both *P. falciparum* and helminths infection reservoirs in Mvomero.

Keywords: Asymptomatic *Plasmodium falciparum*, malaria, Soil transmitted helminths, *Schistosoma haematobium*, Mvomero, Tanzania.

Introduction

Malaria and helminths infections are important parasitic diseases costing developing economies billions of dollars every year (Salim et al. 2015). Sub Saharan Africa currently harbours more than 85% of the estimated global burdens of parasitic diseases (Yapi et al. 2014). The most important helminths infections include *Schistosoma haematobium* and *Schistosoma mansoni* causing urogenital and intestinal schistosomiasis, respectively and the major soil transmitted helminths (STH) including hookworms (*Ancylostoma duodenale* and *Necator americanus), Strongyloides stercoralis*, *Enterobius vermicularis*, *Ascaris lumbricoides* and *Trichuris trichiura*. As a result of geographical overlaps, *Plasmodium falciparum* and helminths share not only the areas in which they occur, but also the human host. Studies suggest that the burden of *P. falciparum* malaria increase with increasing

number of co-infecting helminth species (Kinung'hi et al. 2014, Zeukeng et al. 2014, Dejon-Agobé et al. 2018). It should also be noted that besides having clinical implications (Mwangi et al. 2006, Cooper et al. 2000). *P. falciparum*-helminth co-infections may also complicate control measures disease eradication in endemic areas.

Over more than a decade, several control programs have been put into place to control malaria vectors as well as helminth infections in endemic areas. These include use of insecticide treated nets, indoor residual spraying using pyrethroids and mass drug administration using anthelminthic drugs. However, changes of mosquito feeding and resting behaviour, increased mosquito resistance to pyrethroids (Matiya et al. 2019) and increased prevalence of an outdoor feeding mosquito spp, the *Anopheles arabiensis* (Lwetoijera et al. 2014, Killeen et al 2014) have resulted into increase of residual malaria transmissions in several parts of Tanzania (WHO 2014). This imposes overall challenges to the current malaria vector control measures. Likewise, despite implementation of mass drug administration programs across the country, STH infections continue to persist (Mugono et al. 2014, Bukindu et al. 2016). Meanwhile, the WHO has set targets for global eradication of malaria and STH infections by the years 2020 and 2030, respectively in endemic areas (WHO 2015). In order to achieve the current eradication targets, routine monitoring and evaluating the impacts of the current interventional strategies become critical to inform decision on existing control programs. According to the WHO (2017), routine monitoring and evaluating the impacts of the current interventional strategies form an integral part of preventive chemotherapy programs.

Mvomero is an important sentinel-surveillance-site for both *P. falciparum* malaria and neglected tropical diseases; particularly schistosomiasis and STH infections in Tanzania; therefore important for monitoring effectiveness of the respective control measures. Studies conducted by Mboera et al. (2011) reported over 70% of *P. falciparum* prevalence and *P. falciparum*-helminth (*S. haematobium*, hookworm or *Wuchereria bancrofti*) co-infection rates ranging from 50% to 60% among the school going children in agro-ecosystem communities in Mvomero district Tanzania (Mboera et al. 2011). However, the current status of the burdens of *P. falciparum* infections, STH and *S. haematobium* infections after more than ten years utilization of malaria vector control measures and mass of drug administration using anthelminth drugs in the area has not been established. Therefore, this study aimed at investigating the burdens of asymptomatic *P. falciparum* malaria, helminth and *P. falciparum*-helminths co-infections, and determines the factors associated with asymptomatic malaria-helminth co-infection in the study area. This study forms part of post-control surveillance and is important in informing about the effectiveness of current control programs in the study area.

Materials and Methods

Study area and population

This study was carried out in Mvomero District, Morogoro Region, Tanzania (Figure 1). Mvomero was an ideal site for this study as malaria transmission occurs throughout the year. In that area, temporary and permanent rain puddles as well as seasonal or continuously flooded rice paddies and sugarcane plantations are present. Such environment provides good breeding sites of *Anopheles* mosquitoes and schistosome vectors throughout the year. Apart from schistosomiasis, Mvomero is endemic to other soil transmitted helminths such as hookworms and *Ascaris lumbricoides* (Mboera et al. 2011). This study involved pre-school and primary school-aged children from five wards (Figure 1). The schools included Diongoya and Kaole (urban settings) and Kisala, Mnazi Mmoja, and Mkindo 'A' (rural settings).

Communities surrounding the selected schools are mainly involved in subsistence farming of rice, sugarcane, maize, millet and cassava, and also livestock keeping. The student registration book was used as a sampling frame and study participants were selected using a simple random sampling technique.

Sample size of the study

Sample size for the study was estimated using the following formula described by Pfeiffer (2002):

$$n = Z^2 \text{ P (1-P)/ d}^2$$

where: n = required sample size, Z = multiplier from normal distribution 95% Cl (1.96), P = estimated prevalence 60% of co-infections (Mboera et al. 2011), (1-P) = the probability of having no disease, and d = desired precision (5%).

In this study, the level of confidence set was at 95% (1.96) confidence interval and the prevalence was 60% and 5% set as the precision level for all parameters. Therefore, using the formula, the number of samples obtained was calculated as follows:

$$n = (1.96)2 \text{ x } 0.6\ (1 - 0.6)/\ (0.05)^2$$
$$= 370$$

To account for dropouts from school during the study, 20% of the calculated sample size was added to account for missing samples.

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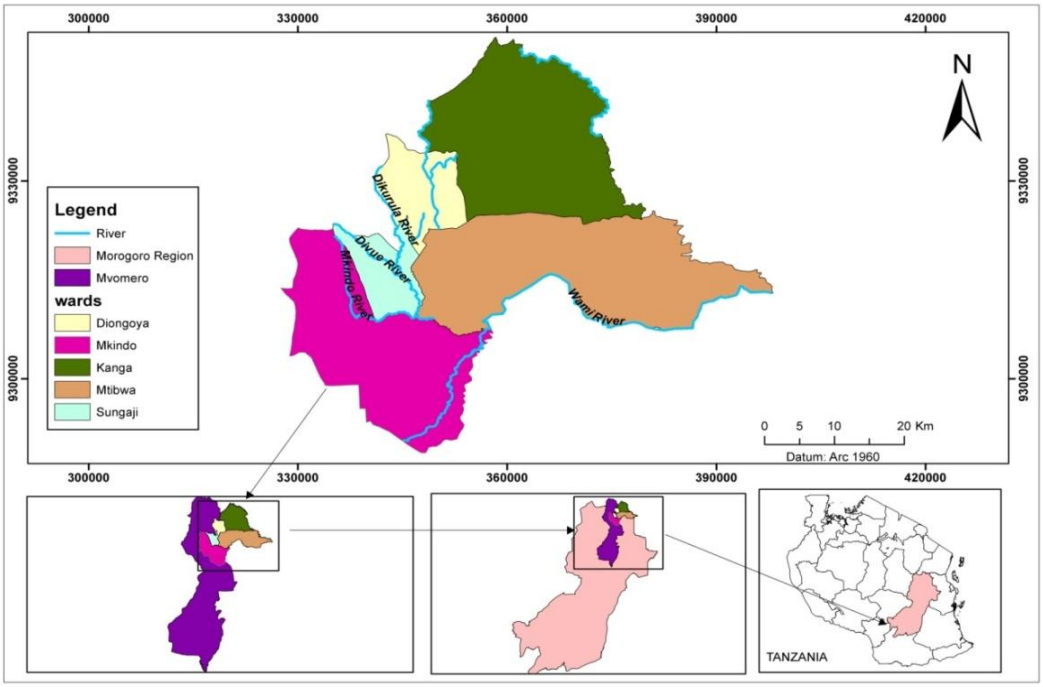

Figure 1: Location of the study area in Mvomero district (Source: Authors).

Study design and data collection

A cross-sectional study was conducted between July and November 2018. Inclusion criteria for the study included children in the 5–16 years of age and children whose parents or guardians were willing to give written consents. Prior to conducting the study, meetings were held with parents/ guardians, teachers and community leaders including village health care workers and village Executive Officers. The aims of the study were thoroughly explained and procedures for data collection were described. Informed written consents were obtained from children

parents or guardians. Finger pricks blood, stool, and urine samples were collected from a total of 374 of children from five schools from July to November 2018.

Demographic data were collected using structured questionnaires. The demographic information collected included age, gender, grade, types of houses living, types of crops cultivating, distance from healthy facility to home and father occupation, insecticide treated net usage and uptake of anthelminth drugs over the past two years.

Parasitological analysis of soil transmitted helminths

Physical appearance of collected stool was recorded e.g., colour, consistency, whether it contained blood, mucus, pus or worms. The stool samples were preserved in 10% formalin. Formal-ether sedimentation technique was used to determine presence of STH in stool samples. Intensity of helminths infections was analysed by MacMaster counter method as described by WHO (1991) and Cheesbrough (2006). Briefly, 7 ml of 5% formaldehyde in saline were added into a mortar. Then, 1 g of stool was emulsified with the formal saline by means of a pestle. The emulsified stool was sieved through a four (4) layers of wet cotton gauge in a funnel into a centrifuge tube. Three (3) ml of ether were then added into a test tube and the mixture shaken for 20 seconds. The contents were then centrifuged at 2,000 rpm for 3 minutes. The fatty coat was dislodged by use of applicator stick. Two thin films of the supernatant were then placed on a microscope slide. One preparation was examined directly, while to the other one, a drop of iodine was added and cover slip placed over before examining under the light microscope using 10x and 40x objectives. Slides smears from centrifuged samples were examined by Mac Master counter slide under 10x and 40x objectives. Identifications of helminths were based on the sizes, shapes and colours of helminths eggs. Egg intensities for intestinal helminths were determined based on the number of eggs detected from each Mac Master counter slide smear. The numbers of eggs detected from each Mac Master counter slides smear were multiplied by 50 to express infection intensities as number of eggs per gram stool (epg). Intestinal helminths egg intensities obtained were classified according to the World Health Organization guidelines (Montresor 1998).

Parasitological analysis of *S. haematobium* infections

Formal-ether sedimentation technique was used to analyse presence of *S. haematobium* in urine samples following the procedures described by WHO (1997) and Cheesbrough (1998). Briefly, 10 ml of collected urine were poured into a conical flask, allowed to sediment for 1 hour, then the supernatant was withdrawn and the sediment transferred into a centrifuge tube and centrifuged at 2000 rpm for 2 minutes. The sediments were examined for the presence of eggs under the light microscope, using x 10 objective. The number of eggs per 10 ml of urine was used to express infection intensity.

Blood sample collection, identification and quantification of *P. falciparum* infections

Finger pick blood was collected for testing malaria infections by a trained laboratory technician. Malaria was diagnosed using microscopy and malaria rapid diagnostic test (mRDT) (SD BIOLINE Malaria Ag *P. falciparum (*HRP2/pLDH-German). Both thick and thin blood smears were prepared for malaria parasite detections. For children that tested positive for malaria with mRDT, their thick blood smears prepared from a finger prick blood were assessed for *P. falciparum* intensity. Slides were stained with field stain air dried for 10 minutes and observed under microscope using oil immersion under 40 x objectives. The number of parasites per microliter of blood was counted against 200 leukocytes (Cheesbrough 2006). The presence of either ring forms or gametocytes was a conclusive

diagnosis of *P. falciparum* infections. Malaria parasite density was estimated by the number of asexual parasites against 200 white blood cells count (WBC) and then multiplying by 40, assuming 8000 WBCs/ul (Cheesbrough 2009). In this study, malaria parasite density, STH and *S. haematob*ium intensity were classified according to WHO (2002).

Assessing behaviour and activities associated with malaria and helminths infections among school going children

Structured questionnaires and oral interviews were administered to assess behaviours and activities that increase risks of malaria-helminths co-infections among children.

Ethical consideration

The study was approved by the University of Dar es Salaam Ethical Committee (UDSM-REC); certificate No **UDSM-REC/2018/02**. Research permit was provided by the Regional Administrative Executive Secretary, Regional Medical Officer in Morogoro and District Educational Officer.

Data analysis

Data were entered, cleaned and validated in the MS-Excel (MS 2010). Statistical analyses were done using IBM SPSS version 24.0 (Armonk, NY: IBM Corp.). Descriptive statistics were used to determine the prevalence of malaria and helminths infections. The arithmetic mean of parasite intensity for each sample was calculated by using the formula by Montresor et al. (1998). That is, Arithmetic mean = $\sum epg/n.$; where: $\sum epg$ = sum of individual *epg*, and n = the number of subjects investigated. Univariate linear regression analysis was used to analyse the associations between intensity and type of helminths infections and asymptomatic malaria parasitemia. Multivariate logistic regression was used to assess the risk factors associated with acquisition of parasites infections. Proportions for categorical variables were compared using chi-square test. Odds ratios (OR) and relative risk (RR) were used to measure strengths of associations between exposures and outcomes. P values less than 0.05 were considered as statistically significant.

Results

Sociodemographic characteristics

A total of 374 primary and pre-school children from five schools were recruited. The mean age of participants was 11.3 years, with an age range of 5 to 16 years. Table 1 shows the characteristics of study participants. Sixty percent, 60% (3/5) of the schools were located in urban areas and 40% (2/5) of the schools were in rural areas. Overall reported Insecticide Treated Nets (ITN) usage was 53%. Recorded school-based uptake of praziquantel for the last 2 years was lower compared to that of albendazole.

Prevalence of asymptomatic *P. falciparum* and helminth *infections*

Prevalence of asymptomatic *P. falciparum* infections is demonstrated in Table 2. Higher prevalence of *P. falciparum* infection was found among children between 11-13 years of age ($t = 9.82$, $p = 0.03$). Most (94.6%) of the asymptomatic *P. falciparum* infections showed a parasitemia not exceeding 500 parasites/ml of blood (Table 1). There was no significant difference between mean parasite density (MPD) between males and females ($t = 6.11$, $p = 0.904$). The mean parasite density was higher among children in 14 - 16 age group (AOR = 1.9, $p = 0.04$). Logistic regression analysis showed children of 14-16 years had higher risks of developing asymptomatic *P. falciparum* malaria with higher parasitemia (AOR = 1.9). There was a moderate positive relationship between the age of the children and *P. falciparum* parasitemia (Pearson correlation, $R^2 = 0.67$, $p = 0.04$)

Schistosoma haematobium was the most prevalent parasite demonstrating 49.7% prevalence (Table 2). Ninety-three point five percent (93.5%) (n = 174) of *S. haematobium* infected children were presented with light egg intensity (1–49 eggs per 10 ml of urine)

whereas 6.5% of the children (n = 12) demonstrated heavy intensity of infections (≥ 50 eggs per 10 ml urine). Out of *S. haematobium*-infected children, girls presented significantly higher mean egg density (MED) than boys ($p < 0.05$) (Table 3). Pearson correlation test showed a weak positive relationship between age of the children and *S. haematobium* infection intensities ($R^2 = 0.219$, $p < 0.001$).

Table 1: Characteristics of study participants

Characteristics	**Percentages (%)**	
Age in years	**Male, %**	**Female, %**
5 -7	67.3	32.7
8 - 10	38.5	61.5
11 - 13	52.2	47.7
14 - 16	58.9	41.1
Parent occupation	**Percentage (%)**	
Farmer	51.3	
Businessman	30.2	
Formal	18.5	
House type	**Percentage (%)**	
Blocks with iron sheet	25.1	
Logs with grasses	42.8	
Blocks with grasses	32.1	
Types of toilet facility	**Percentage (%)**	
Water closet latrine	29.1	
Pit latrine	44.7	
Bush latrine	26.2	
% Uptake albendazole 2017- 2018		
Name of school	**2017, Percentage (%)**	**2018, Percentage (%)**
Mnazi Mmoja	66.3	62.8
Mkindo "A"	80.0	90.1
Diongoya	70.1	74.5
Kisala	76.4	82.7
Kaole	64.2	60.2
Average uptake albendazole	**71.4**	**70.5**
% Uptake praziquantel 2017-2018		
Name of school	**2017, Percentage (%)**	
Mnazi Mmoja	62.8	
Mkindo "A"	79.1	
Diongoya	55.7	
Kisala	72.0	
Kaole	62.3	
Average uptake praziquantel	**66.3**	

Parasite infection burden	**Percentage (%)**
Asymptomatic *P. falciparum* infection	**n = 112**
Light infection	94.6
Moderate infection	5.4
Heavy infection	0
***S. haematobium* infection**	**n = 186**
Light infection	93.5
Heavy infection	6.5
Hookworm-infection	**n = 76**

Light infection	100
Moderate infection	0
Heavy infection	0
***A. lumbricoides* infection**	**n = 47**
Light infection	100
Moderate infection	0
Heavy infection	0

Table 2: Prevalence of asymptomatic *P. falciparum* and helminth infections in relation to children age

Characteristics	**5-7 yrs n (%)**	**8-10 yrs n (%)**	**11-13 yrs n (%)**	**14-16 yrs n (%)**	**Total N (%)**
Plasmodium infections					
P. falciparum (+ve)	11 (22.4)	15 (28.8)	60 (33.7)	26 (27.4)	112 (29.9)
P. falciparum (–ve)	38 (77.6)	37 (71.2)	118 (66.3)	69 (72.6)	262 (70.1)
P. falciparum monoinfection	6 (12.2)	7 (13.5)	42 (23.6)	15 (15.8)	70 (18.7)
***S. haematobium* infection**	18 (36.7)	25 (48.1)	95 (53.4)	48 (50.5)	186 (49.7)
***S. haematobium* monoinfection**	14 (28.6)	20 (38.5)	80 (44.9)	40 (42.1)	154 (41.2)
All STH infections					
Helminth (+ve)	12 (24.5)	17 (32.7)	33 (18.5)	15 (15.8)	77 (20.6)
Helminth (–ve)	37 (75.5)	35 (67.3)	145 (81.5)	80 (84.2)	297 (79.7)
Single STH infections					
Hookworm infection	12 (24.5)	16 (30.8)	34 (19.1)	14 (14.7)	76 (20.3)
Hookworm monoinfection	7 (14.3)	8 (15.4)	21 (11.8)	7 (7.4)	43 (11.5)
A. lumbricoides infection	7 (14.3)	15 (28.8)	15 (8.4)	10 (10.5)	47 (12.6)
A. lumbricoides monoinfection	5 (10.2)	9 (17.3)	11 (6.2)	6 (6.3)	31 (8.3)
Taenia saginata	0 (0)	0 (0)	1 (0.6)	1 (1.1)	02 (0.5)
Schistosoma mansoni	0 (0)	0 (0)	0 (0)	1 (1.1)	01 (0.3)
Mixed helminth infections	3 (6.1)	4 (7.7)	6 (3.4)	3 (3.2)	16 (4.3)
S. haematobium +hookworm	2 (4.1)	0 (0)	4 (0)	2 (2.1)	8 (2.1)
A. lumbricoides + hookworm	1 (2)	2 (3.8)	2 (1.1)	1 (1.1)	6 (1.6)
S. haematobium + A .lumbricoides +hookworm	0 (0)	2 (3.8)	0 (0)	0 (0)	2 (0.5)
***P. falciparum* and helminth co-infections**					
All Plasmodium + helminth coinfection	5 (10.2)	8 (15.4)	18 (10.1)	11 (11.6)	42 (11.2)
P. falciparum + S. haematobium	2 (4.1)	2 (3.8)	9 (5.1)	4 (4.2)	17 (4.5)
P. falciparum +hookworm	2 (4.1)	3 (5.8)	5 (2.8)	2 (2.1)	12 (3.2)
P. falciparum + A. lumbricoides	1 (2)	2 (3.8)	2 (1.1)	3 (3.1)	8 (2.1)
P. falciparum + S. haematobium + hookworm	0 (0)	1 (1.9)	2 (1.1)	2 (2.1)	5 (1.3)

+ve= positive, –ve=negative

Logistic regression analysis showed that children in the 11-13 age group were at higher risks of having higher burdens of *S. haematobium* infection intensities than the rest of the children (AOR = 1.3, p = 0.01, Table 3). Among the STH infections, hookworm was the most prevalent (20.3%) parasitic infection observed in school going children (Table 2). Among the STH infected children, girls had higher mean egg density, although the difference was not statistically significant (t = 4.31, p = 0.705). Pearson correlation test showed a strong negative relationship between age and hookworm egg intensity (R^2 = – 0.73). The multivariate logistic regression analysis showed children in the 8-10 age group had high risks of having more higher hookworm burden (AOR = 1.7, p = 0.03, Table 3).

Table 3: Factors associated with helminth infection intensities in children

Parasites	Covariate	Category	MED	Adjusted OR (95%Cl)	
			Egg/10 ml		**P-value**
S. haematobium			9.3		
	Sex	Boys	11.6	1.0	
		Girls	**13.7**	**1.2 (1.1-5.4)**	**0.02**
	Age group	5-7 yrs	7.4	1.0	
		8-10 yrs	10.5	0.8 (0.2 - 0.8)	0.8
		11-13 yrs	**12.8**	**1.3 (1.2-3.57)**	**0.01**
		14-16 yrs	8.6	0.9 (0.62 -1.19)	0.06
			Egg/gram		
Hookworm			265.68		
	Sex	Boys	256.87	1.0	
		Girls	273.65	0.04 (0.54-0.98)	0.705
	Age group	5-7 yrs	235.43	1.0	
		8-10 yrs	**337.87**	**1.7 (1.2-4.86)**	**0.03**
		11-13 yrs	215.43	1.1 (0.9-3.43)	0.17
		14-16 yrs	207.64	0.04 (0.54-0.84)	0.68
A. lumbricoides			218.33		
	Sex	Boys	198.85	1.0	
		Girls	256.44	0.3(0.24-0.98)	0.06
	Age group	5-7 yrs	278.5	1.0	
		8-10 yrs	252.47	0.04(0.23-0.89)	0.07
		11-13 yrs	204.85	1.82(0.86-3.84)	0.06
		14-16 yrs	180.45	1.67(1.43-3.93)	0.08

***Plasmodium falciparum*-helminths co-infections among school going children**

The overall rate of *P. falciparum*-helminths co-infection (*S. haematobium* or STHs parasites) was 11.2%. High proportions of individuals with asymptomatic *P. falciparum* infections were found to be co-infected with *S. haematobium* (Table 2). Among the *P. falciparum* positive individuals, males showed to harbour higher mean *P. falciparum* parasite density although the difference was not significant (t = 1.45, p = 0.15). Asymptomatic *P. falciparum* parasite density increased with age (Pearson correlation, r = 0.96). *S. haematobium* and hookworm positively influenced asymptomatic *P. falciparum* parasite density (R^2 = 0.55 and 0.73, respectively, Figure 2) among children. Interestingly, a different pattern was observed in in children co-infected with *A. lumbricoides*. In this group, a negative relationship existed between *P.*

falciparum parasitemia and *A. lumbricoides* density (R^2 = 0.02, Figure 2).

Results on bivariate analysis for behaviour and activities of children that increase odds of *S. haematobium*, malaria and STHs infections are stipulated in Table 5. Logistic regression analysis demonstrated that, age of the child, parent being a farmer, involvement of activities in the river or dam, not sleeping under ITN, participating in irrigation scheme (rice or sugarcane farming), doing farm work after school hours particularly preventing birds from picking rice in the field and living in poor houses significantly associated with having asymptomatic malaria-helminths co-infections among children (Table 4). In addition, children of 11-13 years of age were more at a risk of having asymptomatic *P. falciparum* infection-helminths co-infections.

Although prevalence of asymptomatic *P. falciparum* infection was higher in children whose parents were farmers, *P. falciparum* density was found to be similar in all the children studied. Children from Diongoya village had higher prevalence of asymptomatic *P. falciparum* infections (t = 9.8; p = 0.052), however the mean *P. falciparum* parasite density was similar across all the studied villages.

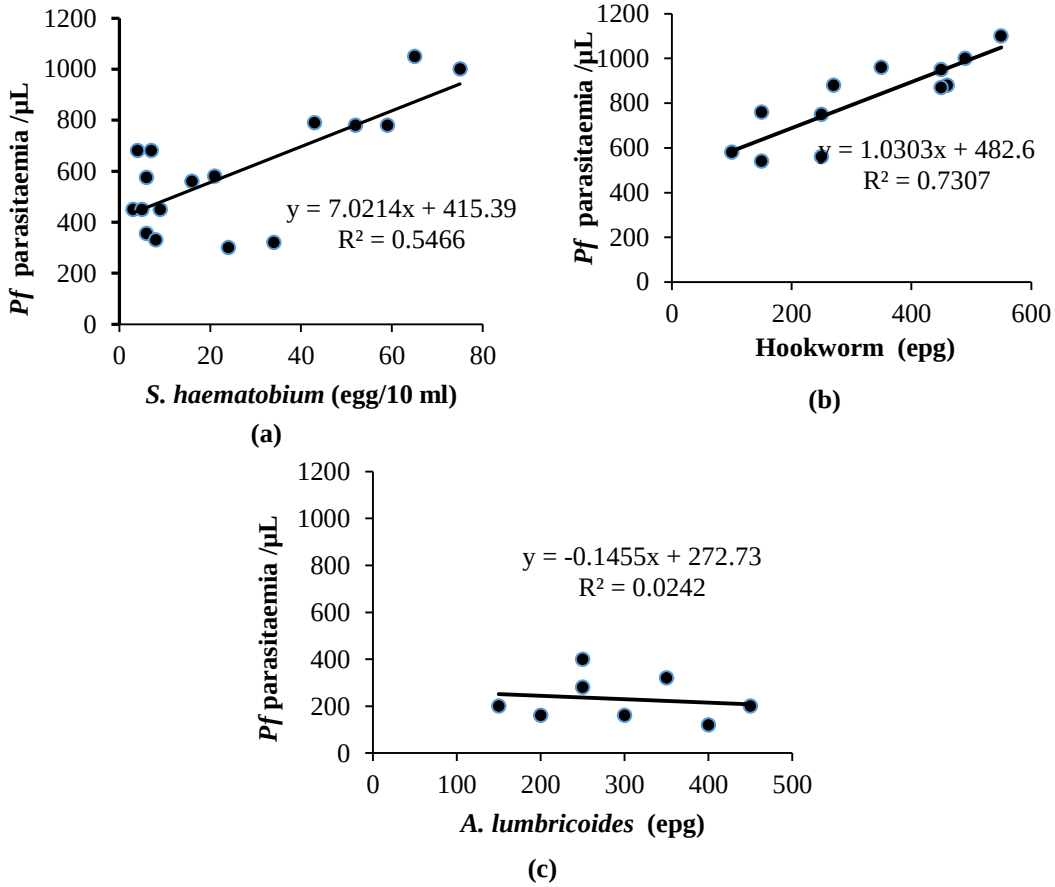

Figure 2: Relationship between mean helminth infections and *P. falciparum* parasite density among *P. falciparum*-helminths co-infected children (a) *P. falciparum–S. haematobium* co-infected; (b) *P. falciparum*–hookworm co-infected; (c) *P. falciparum–A. lumbricoides* coinfected individuals.

Table 4: Factors associated with asymptomatic *P. falciparum* infection-helminth co-infections

Factors	Category	Co-infection		Adjusted OR (95% Cl)	RR	Pearson correlation	P-Value
		YES n = 42 (%)	NO n = 332 (%)				
Age	5-7	05 (11.9)	44 (13.3)	1.0			
	8-10	08 (19)	44 (13.3)	0.67 (0.24-1.83)	0.61	0.292	0.06
	11-13	**18 (42.9)**	**160 (48.2)**	**1.89 (1.22-4.87)**	**2.1**	**0.187**	**0.01**
	14-16	11 (26.2)	84 (25.3)	1.36 (0.87-3.46)	1.2	0.129	0.079
Gender	Male	25 (59.5)	177 (53.3)	1.0			
	Female	17 (40.5)	155 (46.7)	1.28 (0.91-4.72)	1.41	0.083	0.165
Father occupation	Employee	09 (21.5)	60 (18.1)	1.0			
	Businessmen	13 (30.9)	100 (30.1)	1.24 (1.05-5.74)		0.052	0.154
	Farmer	**20 (47.6)**	**172 (51.8)**	**1.75 (1.2-6.83)**	**1.2**	**0.462**	**0.034**
House type	Blocks with iron sheet	10 (23.8)	84 (25.3)	1.0			
	Logs with grasses	**19 45.2)**	**141 (42.5)**	**1.66 (1.23-6.53)**	**1.4**	**0.38**	**0.026**
	Blocks with grasses	13 (31)	107 32.2)	0.46 (0.45-2.68)	0.42	0.027	0.08
Sleeping under ITN	Yes	13 (31)	186 (56)	1.0			
	No	**29 (69)**	**146 (44)**	**1.5 (1.15-4.67)**	**1.83**	**-0.15**	**0.04**
Hand washing behaviour	Yes	10 (23.8)	88 (26.5)	1.0			
	No	32 (76.2)	244 (73.5)	1.2 (0.42-3.65)	1.63	0.062	0.243
Washing fruits before eating	Yes	15 (35.7)	104 (31.3)	1.0			
	No	27 (64.3)	228 (68.7)	1.34 (0.24-4.87)	1.46	0.025	0.074
Types of toilet	Water closet latrine	09 (21.4)	100 (30.1)	1.0			
	Pit Latrine	14 (33.4)	153 (46.1)	1.2 (0.8-2.34)	0.9	0.08	0.06
	Bush Latrine	**19 (45.2)**	**79 (23.8)**	**13 (1.1-4.56)**	**1.2**	**0.3**	**0.01**
Types of crop cultivating	Maize and rice	06 (14.3)	49 (14.8)	1.0.			
	Rice and sugarcane	**18 (42.9)**	**145 (43.7)**	**1.6 (1.21-5.64)**	**1.3**	**0.25**	**0.033**
	Maize and sugarcane	11 (26.2)	81 (24.4)	1.12 (0.82-3.98)	1.41	0.07	0.64
	Millet and maize	07 (16.6)	57 (17.1)	0.8 (0.43-2.76)	0.9	0.068	0.28
Wearing shoes	Yes	14 (33.3)	113 (34)	1.0			
	No	28 (66.7)	219 (66)	1.4 (0.94-3.87)	1.6	-0.063	0.45
Farm work after school hours	Yes	**23 (54.8)**	**165 (49.7)**	**1.96 (1.1-4.63)**	**2.3**	**0.27**	**0.04**
	No	19 (45.2)	167 (50.3)	1.0			
Activities in river/dam	Swimming, fishing	12 (28.6)	97 (29.2)	01			
	Washing, fetching	09 (14.3)	68 (20.5)	0.36 (0.72-2.96)	0.46	0.05	0.3
	Irrigation scheme	21 (50)	167 (50.3)	**1.9 (1.24-5.73)**	**1.84**	**0.143**	**0.02**

Table 5: Behaviour and activities associated with *S. haematobium*, *P. falciparum* and STHs infections

Infection	**Pattern of behaviour/activities**	**Total examined (N = 374)**		**Bivariate analysis**
Soil Transmitted Helminths (STHs)		**STHs (n = 77)**	**No STHs (n = 297)**	**OR (95% Cl)**
	Wearing shoes	10	173	
	Not wearing shoes	67	124	0.1 (0.02-0.6)
	Hand wash after toilet	21	66	
	Not washing hands after toilet	56	231	**1.3 (1.1-2.6)**
	Looking after livestock	18	79	
	Not looking after livestock	59	218	0.87 (0.65-1.2)
	Working in rice field	34	104	
	Not working in rice field	43	193	**1.47 (1.1-3.2)**
Asymptomatic *P. falciparum* malaria		***P. falciparum* (n = 112)**	**No *P. falciparum* (n = 262)**	
	Sleeping under ITN	37	140	
	Not sleeping ITN	75	122	0.4 (0.3-0.9)
	Using mosquito repellents	34	60	
	Not using mosquito repellents	78	202	**1.5 (1.1-2.8)**
	Going to the night ceremonies	69	115	
	Not going	43	147	**2.1 (1.2-4-6)**
	Working in rice field	61	107	
	Not working in rice field	51	155	**1.7 (1.1-3.5)**
	Farming and gardening	47	118	
	Not farming	65	144	0.88 (0.7-1.2)
	Hunting birds	40	133	
	Not hunting	72	129	0.53 (0.4-0.9)
S. haematobium		***S. haematobium* (n = 154)**	**No *S. haematobium* (n = 220)**	
	Crossing river from school	32	45	
	Not crossing the river	122	175	**1.0 (0.6-1.4)**
	Swimming, washing in river and irrigation	66	99	
	Not swimming	88	121	0.9 (90.7-1.4)
	Working in rice field	87	103	
	Not working in rice field	67	117	**1.47 (1.3-3.4)**
	Farming and gardening	68	106	
	Not farming	86	114	0.85 (0.6-1.2)

Discussion

The aim of this study was to assess the current burdens of asymptomatic *P. falciparum* and helminths infections among primary school children in Mvomero district, Tanzania. The results of this study demonstrated that asymptomatic *P. falciparum*, schistosomiasis and STH infections are still prevalent among children in Mvomero. However, there is tremendous reduction of both prevalence rates and intensity of *P. falciparum* asymptomatic parasitemia, STH infections as well as *P. falciparum*-STH co-infections among children in Mvomero compared to the study conducted from 2004 to 2005 by Mboera et al. (2011) in

the same geographical settings. The lower prevalence rates of *P. falciparum* asymptomatic cases and STH infections among school going children in this study may be attributed to the utilization of the current malaria vectors control measures in the country, including the use of ITNs and indoor residual spraying (IRS) and the national-wide mass drug administration using anthelminth drugs. One important finding in this study is the existing high prevalence rate of *S. haematobium* infections (41.2%) among children in Mvomero. The higher prevalence rate of *S. haematobium* entails failure of the current control measures against *S. haematobium* in the study sites.

Although the prevalence of STH infections have gone down, the overall prevalence of 20.6% among school going children is still alarming. Specifically, the prevalence of hookworms' infections recorded in this study is still unacceptably high. The most obvious finding to emerge from the analysis is that low level sanitation was demonstrated by absence of improved toilet facilities to the majority of children. Access to improved toilet facilities is core in the prevention of STH, schistosomiasis and other foodborne and waterborne infections. Over the years, low level of sanitation has been the common occurrence in the STH-affected areas (Gunther and Fink 2010). Consequently, health campaigns as well *wash* interventions; the current WHO strategy to wipe away STH infections (WHO 2017) should actively be integrated with the current anthelminth program in endemic areas. In addition, investment on community information and education programs (Rosemont et al. 1990) is needed particularly those that will help bring changes in behaviour, norms, attitudes and negative perceptions towards STH and schistosome infections.

Comparing these findings with those conducted in the year 2003 by Mboera and colleagues, the prevalence of *S. haematobium* among school going children in Mvomero is still high (Mboera et al. 2011). There are several possible explanations of this result. One is lower praziquantel uptake recorded in this study, which is 12% less than the WHO target for both school-based and community based mass anthelminth treatments. Lower compliance of praziquantel uptake has also been reported in Uganda (Tuhebwe et al. 2015) and Unguja Tanzania (Knopp et al. 2016). Untreated human reservoirs as a result of lower praziquantel uptake may sustain *S. haematobium* transmissions in the study sites. Another reason may be lack of adequate knowledge about the infections and disease among the community members. In addition, the recorded high prevalence of *S. haematobium* among children may be attributed by presence of infected snails vectors, *Bolunus globusus* and *africanus* in the study sites (Mazigo et al. 2012). It is also possible that the increase in prevalence of *S. haematobium* in the study sites is a result of potential existence of reduced efficacy of praziquantel, the current drug used to treat *S. haematobium*. Although there are no reports of drug resistance in the study area, some field and experimental isolates elsewhere have demonstrated reduced susceptibility of *S. haematobium* to praziquantel (Herwaldt et al. 1995, Alonso et al. 2006). Further investigations are required to investigate factors that associate with high prevalence rates of *S. haematobium* infections in the study area to inform decisions on planning effective control strategies. Future studies should also focus on susceptibility status of praziquantel in parasite isolates from different study sites in Tanzania.

In the current study, the presence of co-infecting helminths particularly *S. haematobium* and hookworm in an individual with asymptomatic malaria, significantly increased *P. falciparum* parasite density by 1.2 to 2 folds. This observation could be explained by the fact that, chronic hookworm and *Schistosoma* infections may have induced some levels of T- helper-2 and potentially T-regulatory cells that inhibit T-helper cell-1

responses. T-helper cell 1 responses are critical in clearance of *P. falciparum* infections, and therefore presence of T-helper 2 responses negatively affects control of *P. falciparum* parasitemia. Our study confirms previous established fact that *P. falciparum* co-infections with hookworm and *S. haematobium* may increase the risks of clinical malaria (Zeukeng et al. 2014, Dejon-Agobé et al. 2018). Therefore, according to the present study, *Schistosoma* and hookworm infections may maintain *P. falciparum* parasitemia in individual harbouring asymptomatic malaria in community hence reservoir of *P. falciparum* infections in the community.

Despite the increase in the prevalence of *S. haematobium* infections among school going children, prevalence of asymptomatic *P. falciparum* malaria in the study sites has gone down compared to prevalence rates reported in the past one decade. This is in line with other studies conducted in malaria endemic areas (O'Meara et al. 2008, Carneiro et al. 2010, Winskill et al. 2011, Mawili-Mboumba et al. 2013). The current prevalence of asymptomatic *P. falciparum* malaria is also lower compared to prevalence rates reported by Rumisha et al. (2019) in studies conducted from 2004 to 2005 in Mvomero. The lower prevalence rate of asymptomatic *P. falciparum* parasitemia in this study may be a result of reduction of overall malaria vectors population in the community as a result of utilization of ITN over time; consequently reduction of parasite prevalence hence protection against malaria. Despite the reported lower *P. falciparum* prevalence and intensities in this study, the rate of ITN usage among children is lower (53%) compared to the one reported in the study conducted in the same locality in the past one decade (Rumisha et al. 2019). The WHO's global technical strategy for malaria is to end epidemics of malaria and other neglected tropical diseases by 2030 (WHO 2017). Several questions remain unanswered at present. The important one is: How should the *P. falciparum* asymptomatic reservoirs be dealt with in the malaria endemic areas? Is it about time to institute interventions to eliminate incidence of asymptomatic *P. falciparum* infections in malaria endemic areas? According to Lindblade et al. (2013), asymptomatic *P. falciparum* infections play an important role in malaria transmission. Accordingly, using molecular diagnostic techniques, Lin Ouédraogo et al. (2016) demonstrated that individuals with sub-microscopic *P. falciparum* infections can substantially contribute to onward malaria transmissions in endemic areas.

It should be noted that the prevalence of asymptomatic *P. falciparum* infection in this study is based on microscopically detected *P. falciparum* infections. This might have underestimated the true rates of asymptomatic infections in school going children in Mvomero and hence the actual malaria parasite reservoir pool. Further molecular studies are needed to inform on the true prevalence of asymptomatic malaria in the community. In addition, active malaria case detection and treatment using high throughput methods to detect asymptomatic *P. falciparum* cases in endemic areas will be necessary if malaria elimination goals at year 2030 have to be achieved.

Conclusion

Prevalence of both asymptomatic *P. falciparum* malaria and *P. falciparum*-helminth co-infections has dramatically decreased in Mvomero over the past one decade (from 2004 to 2016). Although prevalence of both asymptomatic *P. falciparum* infections and *P. falciparum*-helminth-co-infections has dramatically decreased in Mvomero over the past one decade, the presence of asymptomatic *P. falciparum* infection carriers may sustain malaria transmission in the study area. High prevalence of *S. haematobium* infections among children in Mvomero implies failure of the current control measures. Integrative control measures incorporating strategies to

combat both helminths and asymptomatic *P. falciparum* reservoirs are important if the WHO 2030-target for elimination of these infections is to be achieved. In addition, more education should be provided to emphasize the uses of ITN among this vulnerable group. Prevalence of *S. haematobium* and hookworm is still alarmingly high. Regular targeted chemotherapy is needed.

Acknowledgments

We are grateful to all the school children who volunteered for the study, teachers and community leaders including Village Health Care Workers and Village Executive Officers for participation and cooperation in this study. We thank Dr. Sixtus J. Assay (a Paediatrician) from Bwagala hospital for scientific advice required for this study. We thank the laboratory technicians Mr. Magnus Mbele (Mhonda TC) and Mwajuma Msuya (Mtibwa hospital) for assistance in laboratory work in the field. Our thanks to members of the Department of Parasitology, Tanzania Veterinary Laboratory Agency (TVLA); particularly Ms. Mary Tarimo and Mr. Charles Ngovi for their support in this study.

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