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Association of Total Levels of Serum Antioxidants with Intensity and Periportal Fibrosis of *Schistosoma mansoni* Infection in Worke Mado, Ethiopia, 2013

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Abstract

Worldwide, soil transmitted helminthes (STH) infections are among the major disease of public health problem in tropical and sub- tropical countries [1]. In many developing countries; one of the most prevalent and most important helminthes are the blood flukes, schistosomes. Schistosomiasis is a chronic illness that can damage internal organs, seriously undermine the work capacity of rural workers and, in children, impair growth and cognitive development. It is caused by several species of which the most prevalent are *S. mansoni*, *S. haematobium*, and *S. japonicum*. It is estimated that 200 million people are already infected of which 180 million are Africans, fewer than 5% of whom are receiving treatment with 779 million peoples at risk. The large fresh water reservoirs associated with ecological changes are contributing for the increased prevalence of *S. mansoni*. The increased prevalence of *S. mansoni* infection results in the formation of periportal fibrosis. Though there are morbidities in different *S. mansoni* areas, disparities in the formation of periportal fibrosis have been observed in different corners of the world. The current study is focused in assessing the prevalence of *S. mansoni*, PPF and measurement of total serum antioxidant (TSA) in *Schistosoma mansoni* endemic and non-endemic areas to associate total serum antioxidants with infection intensity and periportal fibrosis of *S. mansoni* infection in Worke-mado. A community based cross sectional parasitological and schistosomal morbidity survey was conducted on 390 study subjects recruited by systematic sampling technique from the sampling frame in Worke-mado from January to May, 2013. A total of 390 study subjects and 30 controls were included in this study. An overall prevalence of intestinal helminthes in Worke-mado was 76.7% with *S. mansoni* being the dominant parasite comprising 64.4% followed by *A. lumbricoides* (8%). Though not statistically significant, females were more affected than males (35.6% versus 28.7%). Age groups <14 years were more affected than any other groups and was statistically significant. The total prevalence of periportal fibrosis was 9.2% with equal proportion in males and females. The total serum antioxidant (TSA) was 73.1 µmol/L, 70 µmol/L and 305.1 µmol/L in PPF positive, PPF negative and controls respectively. TSA was inversely related with intensity of infection and development of PPF in *S. mansoni* endemic area.

Keywords: *Schistosoma mansoni*; PPF; Ultrasonography; Cross-sectional; TSA

Introduction

Schistosomiasis, also known as bilharziasis, is a chronic illness that can damage internal organs, seriously undermined the work capacity of rural workers and, in children, impair growth and cognitive development (WHO, 2007). Schistosomiasis is a disease of the poor who live in conditions that favor transmission. It is also an insidious disease, poorly recognized at early ages and disabling to men and women during their most productive years and is rarely fatal, but strongly linked to diarrhea, pain, fatigue, hemoglobin deficit, under nutrition, and reduced exercise tolerance (PAHO/WHO, 2007). It is caused by worms (termed flukes), that have a complex life cycle involving freshwater snails as intermediate hosts. Several species exist, of which the most prevalent are *S. mansoni*, *S. haematobium*, and *S. japonicum*. Schistosomiasis is a global parasitic disease which occurs most frequently in sub-Saharan, tropical and sub-tropical areas and ranks second to malaria in terms of socio-economic and public health importance [2].

Schistosomiasis occurs across the African continent in numerous geographic landscapes of varied characteristics, in which specific climatic, physical and human characteristics influence the intensity of transmission [3]. Most transmission sites of *S. mansoni* infections are in agricultural communities along streams between 1300 and 2000 meters altitude infested with *Biomphalaria pfeifferi*, the major snail intermediate host. The prevalence of *S. mansoni* ranges from 10 to 92%. *S. mansoni* transmission above 2200 m and below 800 m is precluded in many parts of Ethiopia by low and high water temperatures, respectively which hinders the growth of the intermediate hosts. In Ethiopia, the optimum range for distribution of *S. mansoni* has been reported as 1500 to 2000 m.a.s.l [4].

The distribution of snail vectors in aquatic environment is increased by accidental airborne transport of snail vectors (Biomphalaria and Bulinus) by aquatic birds. In addition, because snails are hermaphroditic in nature and can self-fertilize then increase in number rapidly, once transported [5]. People are infected with cercaria by contact with water in normal daily activities such as personal or domestic hygiene and swimming, or by professional activities such as fishing, rice cultivation, and irrigation [6]. On contact with humans, the parasite burrows into the skin, matures into another stage (schistosomula), and then migrates to the lungs and liver, where it matures into the adult form. The adult worm then migrates to its preferred body part, depending on its species. These areas include the bladder, rectum, intestines, liver, portal venous

system (the veins that carry blood from the intestines to liver), spleen, and lungs [7]. The adult worm produces huge number of eggs in the intestine and other body sites where many of them are excreted, and the number of eggs per gram of stool detected using kato-katz technique is used for classification of intensity of infection in the community into light, moderate and heavy infections [8]. Substantial numbers of eggs produced by the female worm of *S. mansoni* are also trapped in host tissues such as the liver and intestine. Unlike the adult worms, the trapped eggs induce a local inflammatory response and The-associated cytokines play a prominent role which results in the formation of PPF.

Although estimates of Schistosomiasis indicates the large numbers infected, the disease often receives less attention by health care personnel, national governments and international agencies than it merits. This is partly because not everyone infected will become ill as the degree of morbidity associated with *S. mansoni* infection is related to the intensity of infection [9,10] with heavier infections leading to extensive fibrosis of the liver [11]. It is a disease which remains a major health problem due to lack of vaccines, the failure to eradicate the mollusc vector and the recent development of parasite resistance to anti schistosome drugs [2]. Although the majority of people in endemic areas have only light infections or no symptoms, the impact of Schistosomiasis on economic conditions and the general health situation should not be underestimated (WHO, 2007). Most helminthes infections are prevalent in areas where they are under low economic status with poor access to better nutrition and health care. Likewise, Schistosomiasis is a disease of the poor who live in conditions that favor transmission. It is also an insidious disease, poorly recognized at early ages and disabling to men and women during their most productive years and is rarely fatal, but strongly linked to diarrhea, pain, fatigue, hemoglobin deficit, under nutrition, and reduced exercise tolerance (PAHO/WHO, 2007). Putting in to consideration of the above facts, this study was therefore carried out to assess prevalence and intensity of Schistosoma mansoni, periportal fibrosis and measure total serum antioxidants in Worke-Mado, Northeast Ethiopia with the following specific objective behind.

Methods and Materials

Description of the study area

This particular study was carried out at Worke-mado. It is a village found in Oromia zone of Amhara regional state, 320 km north east of Addis Ababa at an altitude of 1500 m.a.s.l. The people residing in the area are under low socioeconomic and poor environmental sanitation. It is *S. mansoni* endemic area. Addis Ababa is the capital city of Ethiopia and seat for African union and many diplomats and it is found at 9°2'0" North and 38°42'0" East at an altitude of 2200-2500 m.a.s.l. It is non –endemic area for *S. mansoni*.

Study design

Community based Cross sectional parasitological and morbidity survey was conducted in Worke-mado

Sampling technique

Three hundred ninety (390) study subjects were recruited by systematic sampling technique from the sampling frame of the residents of the Kebele and 30 controls were also recruited from Addis Ababa

Sample size determination

The sample size for the cross-sectional survey was calculated using a single proportion formula [12];

$$N = \frac{(Z\alpha/2)2^{*}P(1-P)}{D2}$$ where

N=Desired sample size, Z α/2=1.96 (critical value at 95% confidence level of certainty)

P=prevalence of S.mansoni (65%),

D=Margin of error between the sample and population, 5% marginal error is admitted

$$N = \frac{(1.96)2(0.65)(1-0.35)}{(0.05)2} = 350$$

Taking the assumption of 10% with none and inappropriate responses and adding with the desired sample size calculated above will be 390.

Data management and analysis

Data obtained in the study were analyzed using STATA and association between *S. mansoni* positivity and sex, periportal fibrosis and age categories were computed using chi square (X2) test and odds ratio (OR). Comparison of geometric mean egg count and demographic variables was explored using student's t-test and one-way ANOVA. Partial correlation (r2) analysis was used to assess the relationship between total serum antioxidant and intensity of egg excretion in Periportal fibrosis positives and negative from *S. mansoni* endemic area. Significance was determined at a 5% significance level ($p< 0.05$).

Results

A total of 420, 186(44.3%) male and 234(55.7%) female study subjects were included from Schistosoma endemic and non-endemic areas. Out of which 390 and 30 were from Worke-mado and Addis Ababa respectively. From the 390 total study subjects in Worke-mado, 161(41.3%) and 229 (58.7%) were males and females respectively with a mean age of 17.94(SD=1.5). While the other 30 controls, 25(83.3%) males and 5(16.7%) females with mean age of 27.17 (SD=4.79) were from Addis Ababa. The study subjects age ranged from 8 months to 80 years for Worke-mado comprising of 145 (37.2%), 77(19.7%), and 73(17.8) in the age group of <14, 15-24 and 25-34 respectively. While for the controls, the age ranged from 23-50 where all but one were in the age range of 23-32 years. Among the 390 study subjects from Worke-mado, an overall positivity for at least one parasite was 76.7% with dominant infection being *S. mansoni* (64.4%) followed by *Ascaris lumbricoides* (8.5%) and *Enterobius vermicularis* (2.8%) (Table 1).

Parasite Identified	Total Positive (N%)	Co-infectivity with *S. mansoni* (N%)
Ascaris lumbricoides	33 (11%)	24 (8.0%)

Trichuris trichuria	3 (1%)	3 (1%)
Enterobius vermicularis	11 (3.7%)	8 (2.7%)
Hymenolopis nana	1 (0.3%)	1 (0.3%)
Schistosoma mansoni	251 (84%)	-
Total	299* (100%)	36 (12.1%)
*Percentage is calculated from the total positives (299).		

Table 1: Prevalence of intestinal parasites and co-infectivity with *S. mansoni* in Worke-mado, North east Ethiopia, 2013

As depicted in the above table, more than 12.1% of the study subjects infected with intestinal helminthes were co-infected (double or triple) with *S. mansoni* and other helminthes: the highest co-infectivity being with *A. lumbricoides* (8.0%) followed by *E. vermicularis* (2.7%) and *T. trichuria* (1%) though not statistically significant (P=0.294) (Table 1).

The prevalence of *S. mansoni* in males was found to be lower than females (28.7% versus 35.6%). There was no statistically significant difference (p=0.147) in the positivity of *S. mansoni* between sex though the odds of positivity was 1.4 times higher in females than males (Table 2).

The profile of infection with *S. mansoni* in the various age groups showed a peak infection rate in the age group of <14 years with a prevalence of 28.2% followed by 15-24 and 25-34 years with prevalence of 12.4% and 11.5% respectively; the least positivity being in the age group of >55(2.8%). Prevalence of males in the age group of <14 was higher than females of the same age group (14.6% versus 13.6%). On the contrary, the positivity in the age category of 15-24 was high among females than males (8% versus 4.4%). Likewise, in the age category of 25-34, positivity was high among females than males (8.2% versus 3.3%). There was a statistically significant difference between age category and *S. mansoni* positivity (P=0.002).

In the logistic regression of *S. mansoni* positivity by age category and sex, the odds of *S. mansoni* positivity was 4.2 and 2.8 times higher among age category of <14 years110(43.8%) than the age category of 35-44 (P=0.000) and >55 (P=0.033) years respectively. The least affected to *S. mansoni* infection were in the age category of 35-44 years while the most affected were the age category of 0-14 years. The remaining age categories were not significantly associated with positivity (P > 0.05) (Table 2).

	S. mansoni **positivity (N= male + female)**		**Odds Ratio**	**P value**	**95% CI**
Age Category	<14	110 (43.8%)	1.00	-	Reference
	15-24	48 (19.1%)	0.57	0.066	0.31-1.04
	25-34	45 (17.9%)	0.56	0.066	0.30-1.04
	35-44	18 (7.2%)	0.24	0.000	0.12-0.50
	45-54	19 (7.6%)	0.47	0.068	0.21-1.06
	>55	11 (4.4%)	0.36	0.033	0.14-0.92
Sex	Male	112 (44.6%)	1.00	-	Reference
	female	139 (55.4%)	0.72	0.147	0.46-1.12
251* (100%)					

Table 2: Positivity of *S. mansoni* by Age category and sex in Workemado, Northeast Ethiopia, 2013.

From the total 251positives in Worke-mado, Kato-katz examination revealed a 55% of light infection followed by moderate (29.1%) and heavy infections (16%). Out of the total positives, the intensity of light infection was high among females 91 (36.3%) than males 47 (13.7%). But, moderate and heavy infections were high among males 39 (15.5%) and 26 (10.4%) than females comprising of 34(13.6%) and 14 (5.6%) respectively.

The overall intensity of infection was higher among the age category <14years, 43.8% followed by 15-24years, 19.1% and 25-34 years, 17.9%. The age group of >55 years comprised the least intensity of infection with light (2.8%), moderate (1.2%) and heavy (0.4%) infections and there was a statistically significant difference among age categories and *S. mansoni* infection intensity (P=0.000). The overall intensity of *S. mansoni* among males and females was light (18.7%), moderate (15.5%) and heavy (10.4%) in males and light (36.3%), moderate (13.6%) and heavy (5.6%) in females. There was statistically significant difference between male and female in the intensity of infection (P=0.000). The arithmetic mean (AM) for all study subjects and geometric mean (GM) egg count for the total positives were moderate with 151.3 epg (95% CI, 119.3 -183.2) and 112.79(95% CI, 74.8 - 130.2) epg respectively. The AM and GM egg count of females

was 107.3 epg (moderate) and 89.5 epgs (light) respectively. While for males, it was moderate with 213.8 epg and 150.4 epgs respectively. The geometric mean egg count of the heavily infected individuals was 779 epgs (95%CI, 663.02- 914.88 while it is 203.4 epgs (186.19 - 222.30) and 47.2 epg (95% CI, 43.49 - 51.13) for moderately and lightly infected individuals respectively (Figure 1).

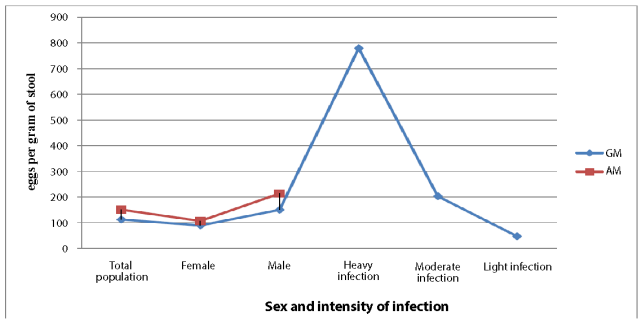

Figure 1: Geometric and arithmetic mean egg by gender and intensity of infection in worke-mado, North Ethiopia, 2013.

The mean egg count was highest among age group of <14 years, 156.96 epg, while the least mean egg count of 57.8 epg was in the age category of 45-54 years. The highest egg count for *S. mansoni* was 3096 epg found in one male participant in the age group of 15-24 years and 2184 epg in females in the same age group. There was a statistically significant difference in mean egg count among males and females (P=0.001) and age categories (P=0.01) (Figure 2 and Table 3).

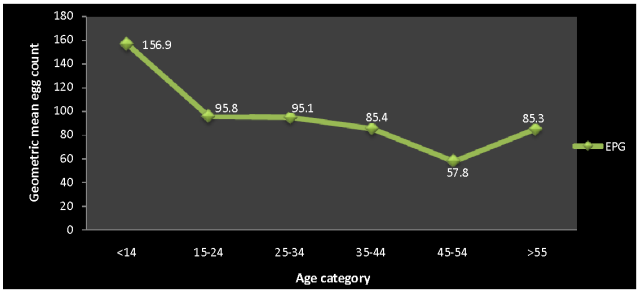

Figure 2: Mean egg count per gram of stool across age category in Worke-mado, North Ethiopia, 2013.

		No of observation	Arithmetic Mean epg	Standard deviation	P value	Geometric mean
Age category	<14	145 (37.2%)	226.76	374.122	0.01	156.9
	15-24	77 (19.7%)	131.22	378.765		95.8
	25-34	73 (18.7%)	128.22	261.969		95.1
	35-44	42 (10.8%)	74.29	171.420		85.4
	45-54	32 (8.2%)	51.00	91.592		57.8
	>55	21 (5.4%)	90.29	225.256		85.3
	Total	390 (100%)	151.26	320.764		112.8
Set	Male	161 (41.3%)	213.8	396.371	0.001	150.4
	Female	229 (58.7%)	107.3	246.158		89.5
	Total	390* (100%)	151.3	320.764		112.8

Table 3: Mean egg count per gram of stool by Age category and sex in Worke mado, Northeast Ethiopia, 2013.

Disparities in development of PPF have been observed in areas with similar *S. mansoni* prevalence. It even has showed higher prevalence of PPF with low prevalence of *S. mansoni* when compared with higher prevalence of *S. mansoni*.

The present study showed that the total prevalence of PPF was 36 (9.2%) with 131 (33.6%) insipient or possible/probable PPF. The prevalence of PPF among males and females was the same (50% each) with no statistically significant difference (P=0.559). There was also no statistically significant difference in the development of PPF among the infection intensities (P=0.057) and positivity (P=0.104) where 8, 20, 3 and 5 of the PPFs were developed in no infection, light, moderate and heavy infections respectively. More than twenty two percent (22.2%) of PPF was developed in individuals with no *S. mansoni* infection and 77.8% of the PPFs were in those who were excreting *S. mansoni* eggs in their feces.

From the total 36 PPF positives 12 (33.3%), 6 (16.7%) and 9 (25%) were in the age category of <14, 15-24 and 25-34 years respectively. The geometric mean egg excretion of PPF positive individuals was 82.71 epg (95%CI, 51.93 - 131.73) while for the border lines it was 123.64 epg (95% CI, 94.46 - 161.83). In multiple logistic regression of age sex, *S. mansoni* intensity, antioxidant level and intestinal nematode in the formation of PPF, only *S. mansoni* intensity was significantly associated (P=0.023, 95%CI 0.99 - 1.00). The odds of PPF development in all age categories except one were almost 1.6 times higher than age category of <14 years with no statistically significant association (P>0.05). Only the age category of 25-34 years was significantly associated (P=0.037) with the development of PPF.

From the total 131 border line PPF individuals 40 (30.5%), 27 (20.6%) and 13 (9.9%) had light, moderate and heavy intensity of

infection. While the PPF positives had 20 (55.6%) light, 3 (8.3%) moderate and 5 (13.9%) heavy infections.

The border line PPF were significantly associated (p=0.027) with intensity of infection. But, the definite PPF positives were not significantly associated with level of intensity of infection (P=0.261).

PPF level	Intensity of Infection					
	No Infection	Light	Moderate	Heavy	Total	P-Value
Normal	80 (35.9%)	78 (35%)	43 (19.3%)	22 (9.9%)	223 (100%)	0.035
Borderline	51 (38.9%)	40 (30.5%)	27 (20.6%)	13 (9.9%)	131 (100%)	0.027
PPF Positive	8 (22.2%)	20 (55.6%)	3 (8.3%)	5 (13.9%)	36 (100%)	0.261

Table 4: Periportal fibrosis in relation to sex and level of intensity in Workemado, Northeast Ethiopia, 2013.

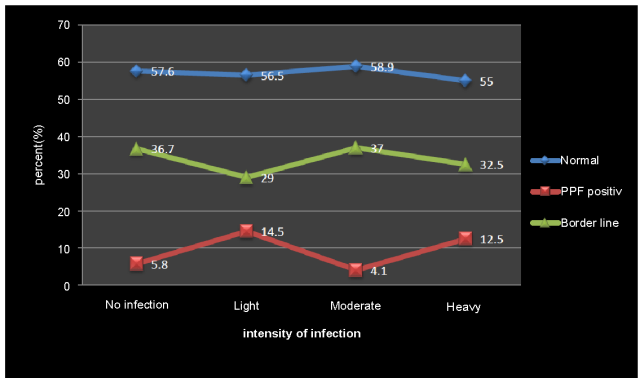

Figure 3: Development of periportal fibrosis in relation to levels of infection intensity.

The risk of periportal fibrosis development in all levels of intensity of infection was almost similar when compared with the non-infected individuals (P>0.05). Similarly, PPF development in females was similar with males (p=0.825) (Figure 3 and Table 4).

After they were assessed by ultrasonography for Schistosoma morbidity, those who had Schistosomal morbidity (periportal fibrosis) and negative volunteered individuals gave 5ml of blood sample for total serum antioxidant measurement. From the 420 study subjects 102 individuals gave blood sample of which 30 were controls from non-endemic area and 72 were from the endemic area. Out of the 72 blood samples collected from endemic area, 36 (50%) were PPF positive while the 36 (50%) were PPF negative.

The mean age of the PPF positive (36) and negative (36) study subjects from Worke-mado were 21.2(SD=4.5) years and 23(SD=5.8) with a mean concentration of the total serum antioxidant (TSA) 73.14 µmol/L (95% CI, 39.25 - 136.29) and 70 µmol/L (95% CI, 53.85- 90.87) respectively. While, the mean concentration of total serum antioxidant of the controls from Addis Ababa with a mean age of 26.9(SD=1.5) was 305.1 µmol/L (95%CI, 255.79 - 363.82) which was much higher than the concentration of both PPF negative and PPF positive participants from *S. mansoni* endemic area. The geometric mean epg of stool in the PPF positive individuals from Worke-mado was 79.3 epg (95% CI, 50.19- 125.14) while for PPF negative and the controls was 172.09 epg (95% CI, 117.48- 252.09) and 0 epg respectively.

PPF status	No of observation below detection level N (%)	No of observation above detection level N (%)	Total N (%)
Positive	26 (72.2)	10 (27.8)	36 (100)
Negative	11 (30.6)	25 (69.4)	36 (100)
Control	0 (0)	30 (100.0)	30 (100)

Table 5: Number of observation in relation to TSA detection level, 2013.

High proportion of PPF positives had serum antioxidant below detection level (<0 µmol/L). On the contrary higher proportion of PPF negative individuals (69.4%) had TSA above detection level. But, all participants from the non-endemic area had TSA above detection level (Table 5).

Individuals who had antioxidant below detectable level (<0 µmol/L) were 12.5 times at higher risk of developing PPF than those who had detectable antioxidant level (>0 µmol/L) and PPF development was significantly associated with detection level of total serum antioxidant (P=0.000).

Total serum antioxidant levels of individuals who had light intensity of infection were 4.4 and 2.6 times higher than those with moderate and heavy infection intensities respectively. It was only with the moderately infected individuals that the antioxidant levels were significantly associated [P=0.006) with PPF development. Although males' antioxidant level was 1.4 times higher than females, it was not statistically significant (P=0.450). TSA was 5.5 and 6.7 times higher in age category of 15-24 and 25-34 respectively than <14 years (P<0.05). There was an inverse relation between concentration of total serum antioxidant and intensity of infection in Periportal fibrosis positives from *S. mansoni* endemic area (r2= -0.22) and PPF negative from

endemic area (r2=-0.43). It was also inversely related with development of periportal fibrosis (r2=-0.45).

Discussion

Worldwide, soil transmitted helminthes (STH) infections are among the major disease of public health problems in tropical and sub-tropical countries [1]. These are closely associated with low house hold income, personal and environmental sanitation and overcrowding, limited access to clean water, tropical climate and low altitude [13,14]. In many developing countries; one of the most prevalent and most important helminthes are the blood flukes, schistosomes [15]. Schistosomiasis is a chronic illness that can damage internal organs, seriously undermined the work capacity of rural workers and, in children, impair growth and cognitive development [16]. Schistosomiasis is an insidious disease, poorly recognized at early ages [16]. Many research have been conducted on intestinal helminthes generally and Schistosomiasis particularly in different corners of the world and Ethiopia with different prevalence ranging from 10%-92%. The results of the present study revealed the occurrence of several intestinal parasites of public health importance. The overall prevalence of intestinal parasite o the current study was 76.7%. This was similar with the research conducted in Waja, southern Tigray, with a total prevalence of 65.6% and Langano SIM and Kime schoolchildren, 83.8%. A nation-wide study on ascariasis in Ethiopia has revealed a low prevalence of ascariasis in the low land and arid areas of the country. In agreement with this report and other reports by Haftay, 201, 2.2%, and Legesse and Erko, 2004, 6.2% this research showed relatively low prevalence of A. lumbricoides (11%). To the contrary, Ascariasis was low as compared with waja (50%). The low prevalence of Ascaris lumbricoides and other parasites other than *S. mansoni* in this area could be due to the fact that it is found in the low lands areas where intestinal parasites are comparatively low as reported by. The prevalence of the intestinal flukes, schistosomes, of the present study showed 64.4%. This is similar with reports from kemissie, 59%, and Bati towns, 51% [17] and endemic sites of Ethiopia, 65.9% [18]. But, it was higher than the studies conducted in shores of Lake Tana, 29%, [19], Wondo Genet, 34.6%, [20], Harbu, 33%, [17], villages of Qalyub region of the Nile Delta, 40.5%, [21], Menofia Governorate in Egypt, 28.5%, and [22]. To the contrary, the results of this study were lower than studies in Tumuga with prevalence of 87% and school children of worke-mado (90%) and sille (95%) [18]. The high prevalence of *S. mansoni* in worke-mado may attribute to the frequent and longer duration of water contact of the residents in the cercaria infested Worke- wenz.

S. mansoni prevalence in males and females (28.7% versus 35.6%) was not significantly different (P>0.05). This was similar with the recent finding in Tumuga where sex related difference in infection was not observed. But, it was higher than the reports from Waja, [17,20] where more males were infected than females.

The profile of infection with *S. mansoni* showed a peak infection rate in the age group of <14 years with a prevalence of 28.2%(<14 years) followed by 15-24 and 25-34 years with prevalence of 12.4% and 11.5% respectively; the least positivity being in the age group of >55(2.8%). The possible reasons for the increased prevalence in age group <14 years could be due to the frequent water contact of children by swimming, fetching water and looking after cattle which gives ample time for water contact, increasing the likelihood of infection. Commonly, once children started contact with water they will have the highest contact time with infected water. Mostly the <14 years age are school children and the location of their school might contribute for the increased prevalence because most of them crosses Worke wonz which will give them ample time for contact with infested water. Besides, Abel et al. [22] reviewed evidence suggesting for individuals having frequent contacts with water infested with the cercaria of S. mansoni, both infection intensities and re-infection after treatment depend, in large part, on their intrinsic susceptibility/resistance to infection, suggesting a role of genetic factors. This age group also covers the highest proportion in the current study (N=145) and this may increased the possibility of having positives [23-25].

On the other hand, other age categories prevalence is reduced due to their maturity and knowledge they have resulting in reduced frequency and duration of water contact and selectivity of swimming, fetching and crossing of the river or water bodies. Besides, in the age group >55, since most of the time they are free from activities that exposes them to cercaria, swimming, fetching and looking after cattle, infection prevalence is reduced. The number of observation of this age group in this study was also few (N=21). In age groups <14 years males were more affected than females and this could be because males spent more time by swimming in the cercaria infected water. To the contrary in other age groups females were more affected than males. And this could be because as they get matured females are engaged in activities that exposed them to cercaria like domestic tasks (washing clothes and house equipments and fetching water) when compared with parallel aged male participants. Besides, as males matured they reduce swimming frequency in the cercaria infected water body and are mostly freed from fetching water. By these and other factors there was a significant difference in *S. mansoni* infection among age categories.

Intensity of infection of this study revealed light (55%), moderate (29.1%) and heavy infections (16%) with moderate geometric mean egg count of 112.8 epg. This was similar with the findings in wondogenet, (Geometric chi eggs per gram of stool=289) [20] and kemissie, Harbu and Bati town with GM egg count of 240, 123 and 195 epg respectively. But, it was higher than Qalyub region of the Nile Delta [21]. The intensity of infection was higher in males (moderate) than females (light) with GM egg count of 150.4 epgs and 89.5 epg respectively(p=0.001). This could be because most of the positives were in the <14 years and in this age group males were frequently exposed to cercaria as explained in the above. Similarly with the positivity of S. mansoni, the intensity was high in the age group of <14 years.

Despite the fact that not everyone infected will become ill as the degree of morbidity associated with *S. mansoni* infection is related to the intensity of infection [9], the disease often receives less attention by health care personnel, national governments and international agencies than it merits. Though only few proportions of individuals develop periportal fibrosis, marked differences in severity of disease have been observed in Africa. The present study showed a total prevalence of PPF, grade 3, 36(9.2%) with 131(33.6%) insipient or possible/probable PPF. The definite PPF were comparable with the research conducted by Nega et al., in worke-mado but twice and ≈15 times that of the research conducted in Sudan [26] and Sille [18] respectively. Whereas, Grade 1and 2 PPFs were similar in Sudan with 33.4%. Besides, there was no significant difference in the formation of periportal fibrosis in gender and levels of infection in the present study. This coincides with research in Sudan [26]. Periportal fibrosis development was homogenous in all age groups except in age group of 25-34 years. This also agreed with the results obtained from Sudan [26]. Many prospects have been suggested regarding the difference of

periportal fibrosis (PPF) development in high and low *S. mansoni* transmission areas in different corners of the world. This might probably be due to a relationship between parasite strain, micronutrients and morbidity in humans. The micronutrients consumed in foodstuffs largely depend on the accessibility and economic status of community in general and individuals in particular. The micronutrients can deter or favor the development of PPF in accordance with the content of their antioxidants. When the pro-oxidant and antioxidant levels are unbalanced, and pro oxidant over weighs, *S. mansoni* morbidity progresses resulting in the development of periportal fibrosis. Alpha (α)-tecopherol and retinol are the major components of anti-oxidant found in serum [18]. Research has been conducted to study the impact of micronutrients by measuring individual components of micronutrients. Assaying each component may not reflect the actual antioxidant level as these components can have synergetic or antagonistic effect on each other. Therefore, the present study was focused on measuring total serum antioxidant (TSA) in Worke-mado.

Study of this research revealed mean concentration of TSA in PPF positives and negatives were 73.14 µmol/L and 70 µmol/L respectively. While for the controls from Addis Ababa it was 305.1 µmol/L. The TSA of healthy controls were more than fourfold of the PPF negative and positive from *S. mansoni* endemic areas. This result agreed with the research conducted by Friis et al. [27], and Pascal et al. [28], which associates *S. mansoni* infection with suboptimal antioxidant micronutrients. This may be because the *S. mansoni* eggs (light and moderate intensity) in infected individuals have an impact in increasing pro-oxidants by producing reactive oxygen species (ROS) that depletes the antioxidant level of individuals. Besides, the health control from Addis Ababa was relatively at better access to antioxidant rich foodstuffs, economic status and safer area for infection when compared with those in *S. mansoni* endemic area. High proportion (72.2%) of PPF positives had serum antioxidant below detection level (<0 µmol/L) while the lower proportion (27.8%) had TSA above detection level. On the contrary higher proportion of PPF negative individuals (69.4%) had TSA above detection level. But, all participants from the non-endemic area had TSA above detection level (Table 5).

Individuals who had antioxidant below detectable level (<0 µmol/L) were 12.5 times at higher risk of developing PPF than those who had detectable antioxidant level (>0 µmol/L) and PPF development was significantly associated with detection level of total serum antioxidant (p=0.000). This agreed with results in the above where higher proportion >72.2% of PPF positives had below detection level and in the PPF negatives and controls higher proportion and all were above detection level respectively. And this indicated that antioxidants levels were predictors of periportal fibrosis development. Total serum antioxidant levels of individuals who had light intensity of infection were 4.4 and 2.6 times higher than those with moderate and heavy infection intensities respectively. In these results though the numbers seem reversed it may be true because there may be other factors (co-infection) that can deplete antioxidants.

TSA was 5.5 and 6.7 times higher in age category of 15-24 and 25-34 respectively than <14 years (P<0.05). This may be because the age category <14 years were intensely infected with 113.7 epg when compared with the rest age categories.

There was an inverse relation between concentration of total serum antioxidant and intensity of *S. mansoni* infection in Periportal fibrosis positives from *S. mansoni* endemic area (r2= -0.22) and PPF negative from endemic area (r2=-0.43). It was also inversely related with development of periportal fibrosis (r2=-0.45). This coincides with findings of (18) in the measurement of serum retinol which is major component of serum antioxidant. This may be because the intensity of infection is associated with increased production of reactive oxygen species (ROS) that deplete the serum antioxidants. As it was explained earlier since the antioxidant levels are predictors of PPF development and intensity of infection are inversely related with antioxidant, the higher the intensity which conversely means the lower antioxidant level (in this research), results in PPF development. But, the antioxidant level was not associated with age either positively or negatively (r2=0.03)

Conclusion

As soil transmitted helminthes (STHs) infection are among the major disease of public health problem in tropical and sub-tropical countries, as of many researches, the present study had revealed a result which is consistent with the truth with a total prevalence of 76.7%. These might closely associated with low house hold income, personal and environmental sanitation and overcrowding, limited access to clean water, tropical climate and low altitude. The current study had revealed *S. mansoni* prevalence of 64.4% and Co-infectivity with other STHs had also been observed. Though there was no significant difference, *S. mansoni* positivity was higher in females than males (35.6% versus 28.7%). The profile of infection with *S. mansoni* had showed a peak infection rate in the age group of <14 years with a prevalence of 28.2% which accounts for their frequent and long duration of contact with infested water. Kato-katz examination revealed a 55% of light infection followed by moderate (29.1%) and heavy infections (16%) and there was a statistically significant difference among age categories and *S. mansoni* infection intensity. There was a statistically significant difference in the GM egg count of females and males with 107.3 epg and 150.4 epgs respectively. Morbidity of S. mansoni, PPF, was seen in 36 (9.2%) of study subjects with no statistically significant difference in males and females (P>0.05). The mean concentration of the total serum antioxidant (TSA) of PPF positive and negative in Worke-mado was 73, 14 µmol/L and 70 µmol/L respectively with a mean concentration of total serum antioxidant of the controls from Addis Ababa, 305.1 µmol/L. And there was an inverse relation with total serum antioxidant and intensity of infection in both PPF positive and negative from worke-mado.

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