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Soil-Transmitted Helminth Infections and Their Associations with Hemoglobin Concentration and Anthropometric Measurements of School Children Injimma Arjo Primary School Children, Oromiya Region, Western Ethiopia

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***Abstracts* –The objective of this study was to determine the prevalence and intensity of major STH infections and their associations with major risk factors, hemoglobin concentration and anthropometric measurements of school children in JimmaArjo primary Schools of Oromia Region. A cross-sectional study was carried from November to January, 2013 in JimmaArjopimary schools and 390 school-children were chosen using stratified random sampling technique and probability proportion to sample size for each class was used. Stool samples of 390 subjects aged 6-17 years were examined using Kato-Katz and formol-ether concentration methods. Direct smear was also employed only for watery stool to detect some intestinal parasites using 0.85% saline solution. 390 blood samples were also collected from the study participants for determination of hemoglobin concentration. Height, weight and body mass index (BMI) of each study participant were measured to determine nutritional status of children as stunted, wasted or underweight. Data were analyzed using the statistical package for social sciences (SPSS version-16) and anthropometry calculating software program (AnthroPlus). The National Centre for Health Statistics (NCHS) growth chart reference was used to determine the proportions of underweight/thinness among 10-17 years old school-children. Hemoglobin level (concentration) was determined using Hemocue HB 201 analyzer. The overall prevalence of STH infection in the school children was 46.7% (182 of 390) and single, double, and triple infections were 32.1, 12.56% and 2.1% respectively. The prevalence of hookworm, A. lumbricoides, T.trichiura, H. nana and E. vermicularis infections were 31.5%, 20.8%, 5.4%, 1.5%, and 0.8%, respectively. The mean egg count of hookworm, A. lumbricoides and T.trichiura were 2198.2±173.1, 2349.12±257.18and 248.7±31.4eggper gram of feaces, respectively. The prevalence of malnutrition in terms of stunting, underweight and wasting was 13%, 17% and 20%, respectively, for those aged from 6-9 years and 39.7% underweight/thinness for those aged from 10-17 years. The overall prevalence of anemia was 13.6%. The findings showed that there was no statistically significant association between STH infections and malnutrition (P>0.05). However, there was significant association between STH infections and anemia (P<0.05). Hookworm infections were considered as causes of anemia among schoolchildren in the study area. Periodically mass drug treatment of school children with appropriate anthelmenthic drug and health education on proper personal and environmental hygiene practices is recommended for school children to keep the prevalence rate of STHs low in the study area.**

***Keywords* – Anemia, Anthropometry, Intensity, Malnutrition, Prevalence, School Children, Soil-Transmitted Helminth. School Children, Soil-Transmitted Helminth, Jimma Arjo.**

I. INTRODUCTION

Helminths are parasitic worms, which infect humans (Maizels and Yazdanbakhsh, 2003), and mainly found in two phyla, Platyhelminths and Nematoda (Hotez*et al*., 2008). Many different species of soil-transmitted helminthes (STHs) infect humans, especially in the tropical and subtropical parts of the developing world. However, four intestinal nematodes in particular stand out because of their widespread prevalence and distribution that result in hundreds of millions of human infections. These include the large roundworm,*Ascarislumbricoides*, the whipworm, *Trichuristrichiura*, and two species of hookworm, *Necatoramericanus* and *Ancylostoma-duodenale (*Hotez*et al*., 2008). These species are collectively known as soil- transmitted helminths (STHs). Multiple infections of these parasites are common in a single individual, especially a child living in a less developed country. Later the infections of STHs in children would cause malnutrition, growth stunting, intellectual retardation and cognitive and educational deficits (WHO, 2005).

In developing countries, including Ethiopia high prevalence of STH infection transmission is attributable to factors associated with low socio-economic status. Such factors include poor personal hygiene and environmental sanitation, with low household income, use of human excreta as fertilizers, lack of shoes, overcrowding and lack of clean water supplies for most parts of the developing countries. In Ethiopia, for example, levels of access to improved sanitation in rural areas are very low (5.4%) (Central Statistical Agency and ORC Macro, 2006). A similar problem prevails in JimmaArjo Town, where the present study was conducted. However, there is no published information in the study area regarding extent and consequences of parasitic helminth infection in children. Therefore, the objective of the present study was toassess the major STH infectionsand their association withhemoglobin concentration and anthropometric measurements of school children in JimmaArjo Primary schools, JimmaArjo Town, West Ethiopia.

II. Materials and Methods

Description of Study Area

The studywas conducted in JimmaArjo Town, in East Wollega Zone of Oromia Regional State, South West Ethiopia. The district capital, JimmaArjo is located at about 379km from Addis Ababa, the national capital and 48 km from Nekemte town the zonal capital. Regarding the altitudinal range, the elevation varies from 1260 to 2520meters above sea level but most areas of JimmaArjo are situated at an altitude greater than 1200 meters above sea level.

According to the 2007 national census (CSA, 2007), the total population of JimmaArjo Town is 86,329, of which 42,093 were males and 44,236 were females within the total area of 75,812 ha. Majority of the population is depends up on the farming of basic crops and animals.The sources of water for home utilization are mainly from wells & springs for urban andrural dwellers. One deep well at Arjo and many on-spot spring system at rural areas provide potable water supply for the community. Available information from the Zonal Water Resource Department indicates that out of the total population in the district, only 7.76 % is supplied with potable water. There is one high school and three elementary schools in JimmaArjo town. The present study was undertaken in JimmaArjoElementary School.

Study Design

The study design was cross-sectional parasitological survey of soil-transmitted helminth infections in primary school children of JimmaArjo Town, West Ethiopia. Prevalence and intensity of soil-transmitted helminth infections of school children in the study area as well as the association of STH infections with anthropometric indices and hemoglobin concentration of school children were determined. The study was conducted from November to January at JimmaArjo primary school, which is found in JimmaArjo Town.

The Study Population

All grades 1-8 students who were at school during the present study were constituted the study population of the present investigation. Only those students who have not taken anthelmenthic drugs for the past three- months, those that were free of malaria infection at the time of sample collection; and those who were willing to participate and produced completed and signed consent form by their parents (caretaker)were included in the present study. Accordingly the study population was390 children inthe JimmaArjo primary school.

Sample Size Determination and Sampling Techniques

The sample size (n)was determined using the statistical formula- $n = (Z\alpha/2)^2 p (1-p)/d^2$ (Kish, 1965). Since the overall prevalence rate (p) of intestinal parasites was not known for the study area, prevalence was taken to be 50%. For the calculation, a 95% confidence interval (z) and a 5% margin of error (d) was used. Therefore, three hundred ninety (390) school-children were chosen to participate in the study. To minimize errors arising from the likelihood of noncompliance, ten percent of the sample size was added to the normal sample.

By using the above formula, the above formula, the study participants were selected using a random sampling technique. The students will be first stratified by stratified cluster sampling techniques according to their educational level (grades 1 to 8). Then, a probability proportionto sample size sampling was under taken for each grade, each class room, and number of sections. Finally, the school children were selected by random sampling technique using random numbers generated from master's lists (roster) of each section documented for 2012/2013 academic year taking as the sample frame.

III. Method of Data Collection

Anthropometric measurements

Body weight and height was measured according to the standardized procedures mentioned in Gibson, (2005) and body mass index (BMI) was calculated using the formula (weight in kg/ [height in m] ^{2}). All the data were transformed and expressed in Z-scores and calculated using anthropometry calculating software program AnthroPlus (WHO, 2007). Under-nutrition was defined for a child, who had less than-2 z-scores (-2SD) from the National Center for Health Statistics (NCHS) median reference population values (WHO, 2007).However, wasting for those children with age above 9 years cannot be evaluated through WHO AnthroPlus, body mass index (BMI) (weight/height in metre^2) was calculated and a BMI-for-age value less than 5th percentile of reference data will be considered as thinness or underweight (WHO, 2009). Data were excluded if a child's height-for-age z-score (HAZ) is below – 6 or above +6, weight-for-age z-score (WAZ) is below – 6 or above +5, weight- for-height z-score (WHZ) is below –5 or above +5, because these extreme values were most likely a result of errors in measurement or data entry (WHO, 2006). Height-for–age, weight-for-age and weight-for- height denote stunting, underweight and wasting, respectively.

Stool sample collection and examination

Formal-ether concentration and Kato-katz techniques were used to assess the overall prevalence of STH infections in the study area. Direct smear wasdone only for watery stool to detect some intestinal parasites using 0.85% saline solution by 10x and 40x microscope objectives.

Blood sample collection and examination

Blood sample was taken by the laboratory technicians from finger pricks of the study subjects to determine the hemoglobin concentration after cleaning the fingers of participants with alcohol on the thumb side of the chosen finger. The collected blood sample was added into a cuvette and placed in HemoCue HB 201 machine. Then the result was read after at least 10 minutes.

Ethical Consideration

Ethicalclearance was obtained from the Medical Ethical Review Committee (MERC) of Nekemte hospital, East Wollega(Appendix- I ,after figures and tables). The permission also obtained from JimmaArjo Town Health Officers, Educational Authorities and School Principals. At the beginning of the study, the objectives and the purpose of the study was explained to the school principal,

parents, guardians and students and the sample was collected from assented children.Individuals diagnosed positive for any Soil-transmitted helminth parasites were treated free of charge with appropriate drug which was provided by the researcher. The prescription on how to use the drug was given by health workers.

IV. Results and Discussion

Prevalence of STH infections among school children

As shown in Table 1, the microscopic stool sample examination showed that infections with various intestinal helminths were common in the study subjects. Of the total 390 children examined, about 46.7% (182 of 390) children were found positive for one or more of intestinal helminths (Table 1). The results obtained in the present study (46.7%) have shown relatively higher prevalence of intestinal helminth infections compared to the results of the studies conducted in school-aged children in Babile town, 27.2 % by Girum (2005) and around Lake Zway, 43.7% by Gezahegn (2008). However, it was lower when compared to study around Lake Zwayisland by Tesfa-Michael and Teklemariam (1983), which showed an overall prevalence of 56.7%. On the other hand, this finding was relatively comparable with other study reported fromHararghe area, eastern Ethiopian region (45.9%) by Ayele and Birre (1989) and south Gonder, 49% by Jemaneh (2000). These differences in prevalence might be a reflection of the difference in local sanitary standard, environmental conditions, time and seasonal differences in the design of the survey work and personal hygiene (Albonico*et al*. 1999).

In this current study, the difference in prevalence of STH infections among school children within the age group and sex was not statistically significant. (Insert table 1. here)

Major soil-transmitted helminth parasites identified in school children

As summarized and presented in the Table 2, five species of intestinal helminths were identified in the study subjects with an overall prevalence of 47.6% (182 of 390 children). The predominant parasites observed were Hookworm (30.8%) followed by *Ascarislumbricoides* (22.1%).Some of the students were found to be infected with *Trichuristrichiura*(5.6%),*Hymenolepis nana* (1.8%) and*Enterobiusvermicularis* (1%)(Table 2). The higher prevalence of hookworm observed in this study might be attributed to the fact that most of the children in the study area do not wear protective shoes, and often they play or go to school and farmlands bare footed. These trends were also reported by Odu*et al.* (2010).

The high prevalence of hookworm in the current study was found to be noticeably higher than that reported in Babile town, 6.7 % (Girum, 2005). However, a similar study conducted in Northwest Ethiopia, Chilga District, showed 37.7% of hookworm infection (Jemaneh, 2001). Variability in endemicity or prevalence of these infections, low sensitivity of the diagnostic method, and the use of single stool sample, environmental contamination and inability of the helminth eggs to withstand diverse temperature could partly explain the observed difference as reported by Mazigo*et al.* (2010). These variations indicate that infection rates depend on such factors as local personal hygienic and sanitary conditions.

*Ascarislumbricoides*was the second most prevalent parasite in this study (Table 2). Its prevalence, 22.1% was found to be higher than the prevalence of *A. lumbricoides* reported in Zway Island, 4.1% by Tesfa-Michael and Teklemariam (1983) and in Lake Langano, 6.2% (Mengistu and Berhanu, 2004). Conversely, its prevalence was lower than the prevalence reported in Northwest Ethiopia, Chilga District, 42.9% by Jemaneh (2001) and different parts of Ethiopia, 37% by Zerihun*et al*. (2008). In contrast, the prevalence of *A. lumbricoides*recorded in the current study was relatively comparable with previous report in Abosa around Lake Zway, 20.5% (Gezahegn, 2008).

The prevalence of *T. trichiura*, 5.4% in the present study was lower than the study conducted in Lake Langano, 14.7% (Mengistu and Berhanu, 2004), Northwest Ethiopia, Chilga District, 14.8% by Leykun, (2001) and different parts of Ethiopia, 30% (Zerihun*et al.*, 2008). The observed differences in the prevalence of intestinal helminth infections in the current study from prevalence of helminth reported in other parts of Ethiopia might be due to differences in diverse environmental conditions of the study sites as epidemiology of soil-transmitted helminths is highly affected by surface temperature (Brooker*et al*., 2003), altitude, soil type, and rainfall (Kariuki*et al.*, 2004). The observed differences could also be explained by the fact that the prevalence and distribution of helminth infections varies by place and with age in Ethiopia as reported by Yeshambel*et al.* (2010).

Among 182 positive individuals, the majority 125(32.1%) of the students had single infection whereas 49 (12.6%) of the students had double infections of any intestinal helminth and 8 (2.1%) of the whole had multiple infections. Therefore, double infections were relatively less common (12.6%) when compared to a similar study that reported 35.4% of double infection rates from southern Ethiopia (Roma and Worku, 1997), however both double and triple infection observed in current study was relatively comparable with the study reported from south Gonder which showed 13.3% and 2.4% double and triple infections respectively, byJemaneh (2000). The most frequent combinations of helminths diagnosed in patients with double infections were *A.lumbricoides and* Hookworm [28(7.2%)] followed by Hookworm and *T.trichuria*. (Insert table 2. here)

Association of intestinal parasitic infections with anemic conditions in school children.

The causes of anemia are known to be complex and multifactorial in origin (Tatala*et al.*, 1998) and several studies have identified iron deficient diet as the primary cause of anemia, while another study indicated malaria as an important cause of anemia (Newton *et al.*, 1997). It is known that inadequate intake of iron is one of the most important factors in iron deficiency anemia (Baynes, 1994). In the present study, emphasis was given to impact of helminthic infections on hemoglobin level of school-

children. About three major intestinal helmiths were observed but only hookworm infected individual showed a significant association with low hemoglobin concentration ($p > 0.05$) (Table 3). This observation is in agreement with the earlier studies done in other parts of Ethiopia (Birmeka, 2007; Woldeyes, 2007; Tsegaye*et al.*, 1999), Malawi (Verhoeff*et al.*, 1998), and in USA (Hawdon and Hotez, 1996). Additionally, similar study done in DebubAchefer District North west Ethiopia (TilahunAlelign, 2010) showed the associations between hookworm parasitic infection and anemia.Hookworm is an important cause of anemia worldwide (Roche and Layrisse, 1966) and this is expressed as a negative correlation between hookworm load and hemoglobin level (Pritchard *et al*., 1991). In contrast to the present observation, Haidar and Pobocik (2009) reported the absence of significant association between hookworm infections and anemia although prevalence of anemia was slightly higher among women aged 15 to 49 years. In addition, a study among schoolchildren in Southeast of Lake Langano, Ethiopia, showed the absence of associations between hookworm infections andhaematocrit values (Legesse and Erko, 2004). Moreover, studies in Panama also failed to demonstrate associations between hookworm infections and anemia at lower intensities (Robertson*et al.*, 1992).The possible explanations for the observed discrepancy between these findings could bewhether or not a person with hookworm infection develops anemia depends on the worm species and load, duration of infection, body iron store, dietary intake and absorption and physiological iron requirements. On the other hand, a low hookworm load can cause anemia in people whose intake of iron is low and whose iron stores are already depleted (Pawlowski*et al*., 1991). In contrast, when high iron intake is available hookworm will not cause anemia even with high worm load. This was demonstrated in children in Botswana by Michaelsen (1985).The present study has therefore, demonstrated a significant association between hook worm infected individuals and low hemoglobin concentration and this indicates that hookworm infection accounts for some of the anemia in this population.

(Insert table 3. here)

Association between Prevalence STH Infections and Anthropometric Indices among 6-9 and 10-17 years Age Groups.

Even though the prevalence of malnutrition was very high among the study subjects, in this cross-sectional study; a significant association was not found between intestinal nematode infections and nutritional status (Table 4). This finding was consistent with previous study, which revealed an independence of anthropometric scores on the overall rate of intestinal helminthic infections (Girum, 2005). Similar results have also been reported from the study conducted in Lowland Bolivia (Tanner *et al*., 2009), in rural Kelantan (Zulkifli*et al*., 2000) and Northern Rwanda (Mupfasoni*et al*., 2009).This could suggest that other factors such as poverty, poor health and sanitary conditions, limited knowledge of nutritional matters among certain households, and fluctuations in incomes which affect the nutritional status are may be predominant among the study subjects as reported by Andrade *et al.* (2001). In addition to this, lack of adequate nutrients caused by high intensity infections in a critical period can prevent the normal growth spurt in pre pubertal and pubertal children (Andrade *et al*., 2001).

(Insert table 4. here)

CONCLUSION

The major intestinal helmiths diagnosed in the school children of JimmaArjo Primary Schools were hookworm, *A.lumbricoides*, *T. trichiura*, *H. nana*and *E. vermicularis.* Hookworm, *A.lumbricoides*and *T.trichuria* were found as dominant parasites in the school-children. The results of this study also showed high prevalence of malnutrition among school children butno association was observed between STH infections and malnutrition as malnutrition is a multifactorialproblem. Hence, detailed studies are required to investigate the association of helminth infection and malnutrition. Even though ,the prevalence of anemia was not a serious problem among the study subjects, because the overall mean hemoglobin value observed in this study (13.52g/dl) was higher than the recommended values, (12 g/dl) (WHO, 1975), there was a significant association between STH infection particularly hookworm infected cases and low hemoglobin concentration. Therefore, high prevalence of hookworm infection in the present study calls for mass drug administration to reduce the prevalence of infection and anemia to below the level of public health importance. Although the pattern found in the assessed potential risk factors data is in the expected direction, there are no statistically significant associations between the most assessed risk factors in the current study and soil transmitted helminth infections.

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AUTHOR'S PROFILE

Gemechis Gemeda Abdissa

The author was born in September, 1991 in Jimma Arjo Woreda, East Wollega Zone of the Oromia Regional State. He started his education at Andinet Elementary and junior secondary School and completed his senior secondary and preparatory education at Arjo Senior secondary high School and at Arjo Preparatory school respectively in Jimma Arjo Town. Upon successful completion of his preparatory school studies, he then joined Wollega University in December, 2009 and graduated with a B.Sc Degree in Applied Biology by scoring very good distinction and winning gold medal in July, 2011. Owing to the merits of his academic excellence, he was selected by Ministry of Education and then he joined Biology Department, postgraduate program, Haramaya University in October, 2011 to pursue his M.Sc. study in Microbiology.

Now I am microbiologist and Lecturer at Jimma University at department of microbiology, biology and also working scientific researches, advising students and other related works of the University ta needed time. I want to do again research mostly on the application of microbiology like industrial microbiology and agricultural microbiology.

My name is Gemechis Gemeda Abdissa , just now ,I would like to express my deepest gratitude to my advisors Dr. Sissay Menkir and Dr.Yitibarek Getachew for their unreserved efforts in advising me to conduct my research and providing me with valuable information and materials. I really appreciate their critical evaluation of the thesis draft and immediate feedback with valuable comments as well as for their immediate response for any of the problems I faced. I am also very grateful to the Jimma Arjo Health Center, for provision of chemicals and materials needed for this study and letting me use all laboratory facilities. Above all, I want to thank the Almighty "God" who strengthened me throughout my career.

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Table 1: Prevalence of intestinal STH infections by age and sex among school children, in JimmaArjo primary school, Oromia Region, Western Ethiopia, 2013

Age group (in year)	No. Examined	Male		Female		Both sex		X^2	P-value
		No. Examined (%)	No. Positive (%)	No. Examined	No. Positive (%)	No. Examined	No. Positive (%)		
6-9	98	55	30(54.5)	43	21(48.8)	98	51(52)	0.939	0.416
10-14	245	118	62(52.54)	127	56(44.1)	245	118(48.16)	1.748	0.202
15-17	47	25	4(16)	22	9(40.9)	47	13(27.66)	0.03	1.00
Allage groups	390	198	96(48.5)	192	86(44.8)	390	182(46.7)	1.73	0.188

Table 2: Major soil-transmitted helminth parasites identified in school children of JimmaArjo primary school ,Oromia Region , Western Ethiopia , 2013

Age Groups (in year)and Sex	No. Examined	Soil- Transmitted Helminths					Multiple infection	
		Al	Hw	Tt	Ev	Hn	Double* infection	Triple** infection
		No.pos (%)	No.pos (%)	No.pos (%)	No.pos (%)	No.pos (%)	No.pos (%)	No.pos (%)
6-9								
Male	55	22(40)	15(27.3)	2(3.6)	-	-	12(21.8)	-
Female	43	12(27.9)	9(20.9)	1(2.3)	-	1(2.3)	4(9.3)	-
10-14								
Male	118	26(22)	41(34.7)	9(7.6)	1(0.01)	4(3.4)	10(0.8)	4(3.4)
Female	127	19(14.96)	42(33.1)	7(5.5)	2(1.6)	2(1.6)	16(12.6)	3(2.4)
15-17								
Male	25	3(12)	4(16)	3(12)	-	-	3(12)	-
Female	22	4(18.2)	9(40.9)	-	1(4.5)	-	4(18.2)	1(4.5)
All age groups								
Male	198	51(25.8)	60(30.3)	14(7.1)	1(0.5)	4(2)	25(12.6)	4(2)
Female	192	35(18.2)	60(31.3)	8(4.2)	3(1.6)	3(1.6)	24(12.5)	4(2.1)
Total	390	86(22.1)	120(30.8)	22(5.6)	4(1)	7(1.8)	49(12.56)	8(2.1)

Al= *Ascarislumbricoides*, Hw= Hookworm, Tt=*Trichuristrichuria*, Ev=*Enterobiusvermicularis*,
Hn=*HymenolpsisnanaEv=Entrobiusvermicularis*,
Double infection* =presence of any two intestinal soil-transmitted Triple infection** = presence of any three intestinal soil-transmitted helminths

Table 3: Association of intestinal parasitic infections with hemoglobin levels and anemic conditions in school children of JimmaArjo elementary school from November-January 2013

STH infections	Frequency (%)	Anemic (%) n=53	Non anemic (%) n=337	OR (95%, CI)	X^2	p-value
Hw						
Yes	120(30.8)	25(20.8)	95(79.2)	0.47(0.25-0.86)	6.07	0.014
No	270(69.2)	28(10.4)	242(89.6)			
Al						
Yes	86(22.1)	16(18.6)	70(81.4)	0.69(0.35-1.36)	1.14	0.29

Table 4: Association of STH infections with anthropometric measurement of school children in JimmaArjo Primary school, 2013

Nutritional Indicators	No. of Examined	Soil Transmitted Helminths		OR (95%,CI)	X^2	P- value
		Positive (%)	Negative (%)			
For 6-9 age Group Stunting	98					
Yes	13(12.3)	8(61.5)	5(38.5)	0.61(0.185-2.02)	0.66	0.42
No	85(86.7)	42(49.4)	43(50.6)			
Underweight						
Yes	17(17.3)	11(64.7)	6(35.3)	0.51(0.171-1.5)	1.5	0.22
No	81(82.7)	39(48.1)	42(51.9)			
Wasting						
Yes	20(20.4)	12(60)	8(40)	0.54(0.19-1.5)	1.37	0.24
No	78(79.6)	38(48.7)	40(51.3)			
For 10-17age group	292	131(44.9)	161(55.1)			
Underweight/thiness						
No(>5$^{th....}$)	176(60.3)	86(48.9)	90(51.1)	1.508(0.94-2.43)	2.86	0.09
Yes(<5$^{th....}$)	116(39.7)	45(38.8)	71(61.2)			

5th…. = 5th percentiles; OR = Odds ratio