




Efficacy of Single Dose Ivermectin Against *Strongyloides stercoralis* Infection Among Primary School Children in Amhara National Regional State

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ABSTRACT

BACKGROUND: Ivermectin has been proven to be highly effective against *Strongyloides stercoralis* in some countries. However, its single dose (200 µg/kg) efficacy has not been proven up until now in Ethiopia.

OBJECTIVE: This study aimed to evaluate the efficacy of single dose ivermectin against *S. stercoralis* infection among school children.

METHODS: Stool sample was collected from April 2019 to December 2019 among 844 school children and screened by formol ether concentration, spontaneous tube sedimentation, Baermann concentration, and agar plate techniques. Single oral dose (200 µg/kg) ivermectin was given to 101 *S. stercoralis*-infected student and posttreatment diagnosis was done for 92 students after 2 weeks.

RESULTS: Of the total 92 *S. stercoralis*-infected students who took ivermectin treatment, 87 were negative with cure rate of 94.6%. No side effect of ivermectin was observed.

CONCLUSION: Single dose ivermectin is an effective dose in uncomplicated chronic strongyloidiasis.

KEYWORDS: *Strongyloides stercoralis*, ivermectin, cure rate, efficacy

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Introduction

Strongyloides stercoralis is a soil-transmitted helminth (STH) that causes strongyloidiasis which is one of the most overlooked diseases among the neglected tropical diseases. Based on estimates using newer diagnostic tools, about 370 million people are infected with *S. stercoralis* worldwide with prevalence rate as high as 60% in endemic areas, especially in Southeast Asia, sub-Saharan Africa, the West Indies and Latin America.¹ Chronic infection (more often asymptomatic) is more common in endemic countries.²

The risk of acquiring *S. stercoralis* infection is higher in rural dwellers, lower socioeconomic groups,³ high open defecation, and low sanitation practice areas.⁴ Skin penetration is the major means of transmission and the filariform larva is an infective stage of the parasite.⁵ Different diagnostic techniques can be used to detect *S. stercoralis*. For instance, formol ether concentration technique (FECT) is a simple diagnostic technique, but has less sensitivity for *S. stercoralis* detection.⁶ The debris is a major factor that obscures detection of *S. stercoralis* larvae by using FECT.⁷ The spontaneous tube sedimentation (STS) technique is also a simple and inexpensive technique that does not require any sophisticated equipment and produces better sensitivity than FECT, but this method is not yet

adopted as routine diagnostic method.^{8,9} The Baermann concentration technique (BCT) is a cumbersome routine diagnostic technique. It has better sensitivity to *S. stercoralis* detection than FECT.¹⁰ Although agar plate technique (APT) is a tiresome routine diagnostic technique, it has a better yield of detection of *S. stercoralis* in the stool.¹¹ The detection rate of APC is 1.6 to 6.0 times more sensitive than FECT.¹² A combination of BCT and APT diagnostic methods increases the detection rate of strongyloidiasis.¹³ Prevention of *S. stercoralis* infection can be enhanced through improved sanitary conditions, wearing of shoes, disposing of feces properly, community education, and prompt treatment of infected cases.¹⁴

Previously, the treatment of choice for *S. stercoralis* infection has been thiabendazole, but this drug is no longer available due to unpleasant side effects. Albendazole, another broad-spectrum antihelmintic agent, was also shown to be effective against strongyloidiasis previously.¹⁵ Both previous and more recent reports show that ivermectin (IVM) is superior to albendazole against intestinal strongyloidiasis. Moreover, a single dose of IVM (200 µg/kg) was shown to be effective in uncomplicated chronic strongyloidiasis.^{16,17} Recently, a preparation of oral IVM licensed for human use has become available in Ethiopia. Nevertheless, albendazole



remains the most widely used antiparasitic drug for the treatment of *S. stercoralis* infection in this country. Therefore, the purpose of this study was to assess the efficacy of a single dose IVM (200 µg/kg) for the treatment of chronic strongyloidiasis in children and to recommend IVM as a means of treatment for strongyloidiasis in endemic countries.

Methods

Study design, area, and period

A cross-sectional study was conducted to determine the IVM efficacy against *S. stercoralis* infection among school children in Amhara National Regional State starting from April 2019 to December 2019. All primary school children aged from 6 to 14 years, living for the last 1 month in the region, volunteer to participate, and give consent or assent were included in the study. Children undertaking anthelmintic drugs for the last 3 months prior to data collection time and those who were not regularly going to school were excluded from the study. Multistage sampling technique was used to select the study areas. The study participants in each class were selected randomly. A total of 844 students were screened for *S. stercoralis*. Those children infected with *S. stercoralis* parasite volunteered to take IVM treatment and gave consent were included in this study. Written informed and verbal consent was obtained from the parents of each child.

Sample size determination

As there is no previous prevalence data conducted at a regional level, the sample size was calculated by take $P=50\%$, 95% confidence interval (CI), 5% ($d=0.05$) margin of error and 2 design effect:

$$n = \frac{Z^2 \cdot P(1-P)}{D^2} = \frac{(1.96)^2 \cdot .5(1-0.5)}{0.05^2} = 384$$

By adding 10% for none response, the total sample size was 422. Design effect (2×422) = 844 study subjects were included in the study. n is the sample size, D is the margin of error, Z is the 95% confidence interval, and P is the prevalence rate.

Laboratory Data Collection

Approximately, 22 g of fresh stool sample was collected from each study participant using a 25-mL stool cup and transported to the nearby health institution to detect *S. stercoralis* using FECT, BCT, STS technique, and APT. No preservative was used during transportation because it might affect the motility and kill the larva of *S. stercoralis*.

In FECT, approximately, 0.5 g of stool sample using plastic stick was transferred to the sample collection tube. The cover of the sample collection tube was discarded; the filtration concentration unit with conical tube was introduced and screwed. The sample was mixed up carefully, turned over, and spun at 1000g, for 3 minutes. The supernatant was removed and the sediment

was put on the slide and looked for *S. stercoralis* larvae using microscope.¹⁸

In the STS, around 3 g of stool sample was weighed, homogenized in 10 mL of saline, filtered through surgical gauze into a 50-mL plastic tube, and then filled with more saline solution. The tubes were plugged and shook vigorously and left to stand for 45 minutes. Finally, the supernatant was descanted and a sample was taken from the bottom and put on a slide. The slide was observed using a microscope to check the presence of larvae of *S. stercoralis*.⁹

In the APC, about 3 g of feces was placed on the center of a nutrient agar plate in a Petri dish. The Petri dish was sealed with adhesive tape and incubated at 26°C for 2 days or for 48 hours. The adhesive tape was removed; 5 mL of 10% formalin was added to the agar surface, waited for 5 minutes, transferred to a conical test tube, and centrifuged at 1500 for 5 minutes. Finally, the sediment was first looked for *S. stercoralis* with a microscope.¹²

In BCT, approximately 15 g of fresh, unrefrigerated stool sample was weighted and mixed with water and charcoal powder, transferred to a Petri dish covered with paper towel and incubated for 24 hours at 26°C. The incubated stool sample was suspended in a funnel connected with a rubber tube which contains warm water for an hour. The filtrate in the rubber tube was collected with 15 mL conical test tubes and centrifuged for 5 minutes at 2000 r/min. The supernatant was discarded and the sediments were mixed and seen with a microscope.¹⁹

Those school children who were found to be positive for *S. stercoralis* using any one of the above diagnostic tests were considered as positive and became a candidate for evaluating the efficacy of IVM treatment.

Evaluating the Efficacy of IVM

Training on *S. stercoralis* transmission and prevention was given to 101 *S. stercoralis*-infected school children. The children were instructed to wear shoes when walking, going to latrine, staying on playing grounds, and playing with soil. Moreover, they were instructed not to get involved in irrigation activities for 2 weeks. Then, we communicated all *S. stercoralis* positive cases to be volunteered to give stool samples after 2 weeks of treatment. Strongyloidiasis-positive students who got proper training were recruited and treated with IVM or *Mectizan* (Manufactured by Merck & Co., Inc., Paris, France with 3 mg preparation). *Strongyloides stercoralis*-infected children was treated with single dose oral IVM at 200 µg/kg/d at day 0.²⁰ Ivermectin-treated school children were followed up for 2 weeks for allergy, wearing shoes regularly and their sanitation condition by school teachers and health extension workers. Stool samples were collected from children who took IVM treatment after 2 weeks and then tested by BCT, STS, and APT for *S. stercoralis*. The percentage of strongyloidiasis cure rate (CR) induced by the treatment was calculated as a ratio of the number of children who were negative for *S. stercoralis* larvae after treatment to the number of *S. stercoralis* positive children before treatment.²¹

Table 1. Sociodemographic variables and distribution of intestinal parasites among primary schools students.

VARIABLES	TOTAL EXAMINED NO. (%)	TOTAL POSITIVE NO. (%)	NEGATIVE NO. (%)	χ^2 , P VALUE
Age, y				
6-9	254 (30.1)	169 (66.5)	85 (33.5)	6.60, .04
10-11	364 (43.1)	254 (69.8)	110 (30.2)	
12-14	226 (26.8)	174 (77.0)	52 (33.0)	
Sex				
M	436 (51.7)	323 (74.1)	113 (25.9)	4.88, .03
F	408 (48.3)	274 (67.2)	134 (32.8)	
Residence				
Urban	99 (11.7)	66 (65.2)	33 (34.8)	0.90, .35
Rural	745 (88.3)	531 (71.0)	214 (29.0)	
Total	844 (100)	597 (70.7)	247 (29.3)	

“Cure” is defined as the absence of *S. stercoralis* larvae in the stool at day 14 of treatment. “Failure” is defined as the presence of larvae 2 weeks after initiation of treatment:

$$CR = \frac{\text{The number of study subjects who was negative after treatment at day 14}}{\text{The total number of positive participants}} \times 100$$

To ensure reliable data collection, training of laboratory personnel on sample collection, diagnosis, and explanation about the study were given prior to stool sample collection. Proper labeling of the stool cup with serial numbers was done. The amount of stool sample was checked during collection. Stool samples were transported to the nearby health institution as soon as possible. Standard operating procedures (SOPs) and accuracy of test results were maintained. Regular supervision by principal investigator was done. To eliminate observer bias, stool slides were examined independently with 2 laboratory personnel and the results of their observations were recorded for later comparison on separate sheets. The discordant results were checked by the principal investigator. Generally, the quality assurance was checked during preanalytical, analytical, and postanalytical stages.

Statistical Data Analysis

Data were entered and analyzed using Statistical Package for Social Sciences (SPSS) version 23 statistical software. First, the overall prevalence of *S. stercoralis* was calculated by descriptive statistics and chi-square. And then *S. stercoralis*-infected children were treated with IVM and the CR of IVM was calculated using descriptive statistics at 95% CI.

Ethical clearance was obtained from the Ethical Review Committee of Science College, Bahir Dar University, and the Amhara Regional Health Bureau. Permission letters were also secured from Amhara Regional Education Bureau, Zonal and Woreda Education Offices. Written informed consent was obtained from the parents/guardians after explaining the purpose and objective of the study. Enrollment in the study was purely on voluntary basis and the study participants' laboratory results were kept confidential. Study participants who were positive for any other intestinal parasites other than those participated in the study of the efficacy of IVM were referred to the nearby health institutions for treatment. School children who felt discomfort for any reason to give stool samples after 2 weeks were withdrawn.

Results

Sociodemographic characteristics of the study participants

A total of 844 primary school children were included in our study. The children's mean age was 10.3 years (age range: 6-14 years) with an SD of 1.77. Male students accounted for 51.7% and most (88.3%) were rural dwellers (Table 1).

The overall prevalence of intestinal parasitosis with a combination of FECT, STS, BCT, and APT was 594 (70.7%). The 12- to 14-year age group had the highest prevalence (77%) (Table 2). The total prevalence of STHs accounted 311 (36.9%). Parasites identified were 277 (32.8%) hookworm species, 201 (23.8%) *Entamoeba histolytica/dispar*, 170 (20.1%) *Schistosoma mansoni*, 127 (15.1%) *S. stercoralis*, 62 (7.4%) *Giardia lamblia*, 37 (4.4%) *Ascaris lumbricoides*, 33 (3.9%) *Hymenolepis nana*, 7 (0.8%) *Enterobius vermicularis*, 6 (0.7%) *Trichuris trichiura*, 4 (0.5%) *Taenia* species, and 3 (0.4%) *Fasciola hepatica*.

Table 2. The distribution of *Strongyloides stercoralis* among school children with their sociodemographic variables.

VARIABLES	TOTAL EXAMINED NO. (%)	S STERCORALIS POSITIVE, NO. (%)	S STERCORALIS NEGATIVE, NO. (%)	χ^2 , P VALUE
Age, y				
6-9	254 (30.1)	24 (9.4)	230 (90.6)	9.04, .01
10-11	364 (43.1)	62 (17.0)	302 (83.0)	
12-14	226 (26.8)	41 (18.1)	185 (81.9)	
Sex				
M	436 (51.7)	69 (15.8)	367 (84.2)	0.43, .56
F	408 (48.3)	58 (14.2)	350 (85.8)	
Residence				
Urban	99 (11.7)	13 (13.1)	86 (86.9)	0.32, 0.66
Rural	745 (88.3)	114 (15.3)	631 (84.7)	
Total	844 (100)	127 (15.0)	717 (85.0)	

Table 3. Efficacy of ivermectin against *Strongyloides stercoralis* infection by different diagnostic methods.

DIAGNOSTIC METHODS	S STERCORALIS POSITIVE RESULT, NO.	IVM TREATED, NO.	STOOL GIVEN POST TREATMENT, NO.	POSTTREATMENT S STERCORALIS RESULT, NO.		CR (95% CI), %
				NEG	POS	
FECT	17	14	NA	NA	NA	NA
STS	34	27	23	20	3	87.0 (67.9-95.5)
BCT	86	65	65	60	5	92.3 (83.2-96.8)
APT	91	68	68	63	5	92.7 (83.9-96.8)
Combination of all methods	127	101	92	87	5	94.6 (87.9-97.7)

Abbreviations: APT, Agar plate technique; BCT, Baermann concentration technique; CI, confidence interval; CR, cure rate; FECT, formol ether concentration technique; IVM, ivermectin; STS, spontaneous tube sedimentation.

Higher prevalence of *S. stercoralis* was recorded among school children in the age group 12 to 14 years (18.1%), boys (15.8%), and rural dwellers (15.3%) (Table 2).

Efficacy of IVM Against *Strongyloides stercoralis*

A total of 127 school children were infected with *S. stercoralis*. Of which, 92 (72.4%) *S. stercoralis*-infected school children were co-infected with other parasites. From 127 *S. stercoralis*-infected school children, 101 children volunteered to take IVM treatment. After 2 weeks, 92 students gave stool sample for re-checkup and 87 *S. stercoralis*-infected children were found to be cured. The CR of IVM against *S. stercoralis* was 94.6% (95% CI: 87.9%-97.7%). Five (5.4%) students were positive for strongyloidiasis post treatment by BCT and APT methods and only 3 (3.3%) of *S. stercoralis*-infected students were positive by STS after treatment. Better CR rate was obtained by combination of methods (Table 3). Those *S. stercoralis*-positive children after single dose IVM treatment showed a decreased larval number count in the posttreatment detection. During

the 14 days posttreatment follow-up, no side effect and allergic reaction was observed among students who took IVM treatment. There were 9 children who lost their follow-up due to absentee during posttreatment data collection period.

Discussion

Strongyloidiasis is one of the neglected tropical diseases which is common in tropics and subtropics. The total prevalence of *S. stercoralis* in the present study was 15% (95% CI: 12.8%-17.6%). This finding is consistent with earlier prevalence (15.9%) study conducted in Thailand,²² but it is lower than previous report of 27.7% in Brazil,²³ 24.4% recorded in Cambodia,²⁴ and 20.7% in Bahir Dar, Ethiopia.²⁵ The current result is also high than earlier prevalence obtained (5.8%) in Southeast of Lake Langano, Ethiopia,²⁶ 10% in Angola.²⁷ The prevalence difference might be due to the difference in the detection methods used. For instance, a study conducted in Bahir Dar, Ethiopia, used molecular detection method which has high sensitivity in combination with FECT and BCT.²⁸ As

a result, better detection rate was obtained. On the other hand, a study conducted in Southeast of Lake Langano used Kato Katz and FECT which have relatively low detection rates.²⁹

The first-line drug for *S. stercoralis* is IVM. The CR of IVM (94.6%) obtained in the present study was comparable with CR previously reported 95.2% in Switzerland,³⁰ 96.8% in Thailand,²⁰ 98.7% in Thailand,²² 98.3% in Cambodia,²⁴ and 96% in Japan.³¹ The efficacy obtained in the present was also higher than earlier 85.7% efficacy report in Italy,³² 84.07% in Nigeria.³³ The lower CR or the presence of *S. stercoralis* larvae after 2 weeks of post treatment in other previous studies might be due to resistance of IVM or cross-resistance among macrocyclic lactones.³⁴ It might also be ineffective action of IVM against the larvae and eggs of *S. stercoralis* in the human tissues. In addition, it might also be due to the autoinfection cycle which is completed in 3 to 4 weeks in human tissue and poor immune system of individuals.³⁵

In the present study, there was no any adverse reaction observed among participants who took IVM treatment. This result was consistent with previous reports obtained from a clinical trial conducted in Thailand.²⁰ Our present results are in line with the findings of previous researchers in which they showed that IVM is safe and has minimal side effects.^{16,17}

Generally, the current report clearly showed the efficacy, the advantage of single oral dose treatment, and absence of adverse effects with IVM treatment against strongyloidiasis.

Conclusion

In this study, single dose (200 µg/kg) IVM is a better treatment regimen and no adverse reaction is recorded. Therefore, single dose IVM should become an alternative treatment of choice for uncomplicated chronic strongyloidiasis, especially in a developing country including Ethiopia, where IVM is not currently used as a treatment against *S. stercoralis* infection.

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Author Contributions

TH designed the study, developed the proposal, collected the data, and critically revised the manuscript. EN designed the study, critically followed the data collection and laboratory diagnosis, and revised the manuscript. AA designed the study, critically followed the data collection and laboratory diagnosis, and revised the manuscript. AM critically revised the manuscript. MA entered and cleared the data.

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