EFFICACY OF ALBENDAZOLE AND RE-INFECTION RATES OF SOIL-TRANSMITTED HELMINTHS AMONG SCHOOL CHILDREN IN JIMMA TOWN, SOUTHWEST, ETHIOPIA

BY: - ALEMU WORKU (BSc.)

A THESIS SUBMITTED TO SCHOOL OF MEDICAL LABORATORY SCIENCES, FACULTY OF HEALTH SCIENCES, INSTITUTE OF HEALTH, JIMMA UNIVERSITY IN PARTIAL FULFILMENT OF THE REQUIRMENTS FOR THE DEGREE OF MASTER OF SCIENCE IN MEDICAL PARASITOLOGY

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ABSTRACT

Background:- *Schoolchildren (SC) are at higher risk of infection with soil-transmitted helminths (STHs) throughout the world. The STHs remain major public health problem in the poorest communities causing poor school performance and reduced productivity. In Ethiopia, STHs infections in SC remain high despite the fact that periodic administrations of anthelmintic drugs have been underway for the last five years. Moreover, little information is available regarding re-infection rates & current efficacy of albendazole (ALB) among SC at national and local level*

Objectives:- *To assess efficacy of single dose (400mg) ALB against STHs infection and re - infection rates of STHs among SC in Jimma Town, Southwest Ethiopia*

Methods: - *Prospective cohort study design was employed involving 393-SC attending 4 primary schools in Jimma Town between April to September 2017. Socio-demographic data was collected using semi-structured questionnaire. Stool specimens were collected at baseline, after 3 and 18- weeks post treatment. To express infection intensities; stool sample examined using double Kato Katz smear technique and quantify the number of eggs per gram (EPG). The efficacy of 400mg ALB and re-infection rates of STHs was determined after three and eighteen weeks respectively.*

Results: - *The overall prevalence of STHs infection at baseline was 35.1%. The prevalence of A. lumbricoides, T.trichiura and hookworm infection were 14.5%, 22.4% and 5.3%, respectively. After 3 weeks post-treatment higher efficacy of ALB found on A. lumbricoides (ERR=99.9%) followed by low efficacy against hookworm (ERR=54.4%) and T. trichiura (ERR=38%). The overall prevalence of the three STHs at 18-weeks post-treatment was 23.4% and the re –infection rates was 26.3%. Re -infection rates of A. lumbricoides was 14.5%; whereas re infection rates of T. trichiura and hookworm was 27% and 19%, respectively.*

Conclusions and recommendation: - *There is a high re-infection rate of STHs and poor efficacy of 400mg ALB against T. trichiura and hookworm infection among SC in our study area. Strong and integrated prevention and control method is mandatory to alleviate the problem.*

Key words:-*Soil-transmitted helminths, Reinfection, Efficacy, Schoolchildren, and Albendazole*

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ALB	Albendazole
CR	Cure rate
EPG	Egg per Gram of stool
ERR	Egg Reduction Rates
IPs	Intestinal parasites
MDA	Mass Drug Administration
PSAC	Preschool-aged children
SC	Schoolchildren
SOPs	Standard operating procedure
SSA	Sub-Saharan Africa
STHs	Soil-transmitted helminths
WHO	World Health Organization

1. INTRODUCTION

1.1. Background

Soil-transmitted helminths are a group of nematodes that infect more than a billion people worldwide(1, 2). The four main STHs species that infect people are the roundworm (*Ascaris lumbricoides, A. lumbricoides)*, the whipworm *(Trichuris trichiura, T. trichiura) and* two hookworm species (*Ancylostoma duodenale* and *Necator americanus*). The highest number of STHs infection occurs in Central and South America, People's Republic of China, Southeast Asia, and Sub-Saharan Africa (SSA)(3, 4). In 2010, more than 1.4 billion people were infected with at least one of the three STHs, resulting in a global burden of approximately 5.2 million disability-adjusted life-years (DALYs) (20% of total number of DALYs attributable to Neglected Tropical Disease (NTDs)(5).

The occurrence and distribution of STH infection are directly associated with socio economic status of the communities; poor sanitary facilities; improper disposal of human excreta, insufficient water, sanitation, and hygiene (WASH); climatic characteristics of the area and substandard housing and lack of education(6, 7). Occurrence of infection is also associated with associated other factors like, ignorance of simple health promoting factors and overcrowding, limited access to clean water, tropical climate and low altitude(8).

People get infected by these parasites through direct penetration of the skin or ingestion of eggs developed in the contaminated soil in areas where sanitation status is poor. Before becoming adults in their human host the larvae of hookworm and *A. lumbricoides* migrate through the heart and lungs for ten days during which time the larvae grow and develop(9, 10). Preschool-aged children (PSAC), SC and women of child-bearing age are the segments of people at especial risk of STHs morbidities (11). According to World Bank ranking, STH infection causes more ill health in SC age b/n 5-15 years than any other infection(12). Untreated, chronically infected children might suffer from anemia, malnutrition and impairments in cognitive and physical development impacting their school attendance and performance (2).

Ethiopia; one of the developing country in Africa is suffers from high burden of hookworm *infection, A. lumbricoides* and *T. trichiura* (4, 13). Numerous studies indicated that because of

Ethiopia has one of the lowest quality drinking water supply and latrine coverage in the world, hence, STH is widespread and cause significant morbidity in the country(14). Most parts of Ethiopia are suitable for the transmission of STH infection; except parts of Somali and Afar regions where the annual mean temperature is too high for transmission(15). Large scale distribution of anthelminthic drugs without prior diagnosis (preventive chemotherapy) recommended by the WHO mainly given to SC, is the current strategy to control or reduce morbidity(16). As major means of controlling these STHs infection the periodic administration (MDA) recommended by the WHO is either single dose of ALB or Mebendazole (MBD). Albendazole, the most widely used anthelmintic drug against *A lumbricoides* and hookworm, is highly effective, both in terms of CR and ERR, while mebendazole is the second most widely used drug for infections with STHs(3, 17).

Environmental control of STH specifically improving access to safe water supply, basic sanitation and improved hygiene may serve to complement this deworming effort and proven to be cost effective(18). However, the main problem and challenge to control of STH is re infection rate and low efficacy of both ALB and MBZ especially against *T. Trichiura*. Even though STH re infection anticipated being occurring rapidly post-treatment with the standard anthelminthic drugs in endemic settings like Ethiopia, because there is a lack of information regarding the re-infection status and level in susceptible population.

The efficacy of ALB in terms of ERR usually measured using qualitative and quantitative diagnostic tests for eggs or larvae in feces at an optimal time interval post-treatment, which is dependent upon the species of each parasite. Several studies reported that ALB is effective against *A. lumbricoides* and hookworm at a single dose administration. However, it is less effective against *T. trichiura* using the same dosage administration as compared to against *A. lumbricoides* and hookworm with a slight difference in Cure rate (CR) and eggs reduction rate (ERR) from studies to studies. Cure rate and ERR are indicators commonly used to measure the reduction in prevalence and reduction in intensity of infection.

There are different diagnostic techniques such as (FLOTAC, McMaster, and Kato Katz thick smear) employed to monitor the efficacy of ALB against STH in terms of ERR by different researchers in different areas of the world, each having its own weakness and strengths. After

deworming STH with 400mg ALB, many studies measure the efficacy ALB by counting fecal ERR and CR. However, ERR is considered as more appropriate standard and the best indicator for measuring the efficacy of ALB than CRs in highly endemic areas(19).

It has been underlined that, it requires continuous assessment of evidence based information about the re infection rates of STH and assessing regularly the efficacy of ALB in different settings to provided future direction on the strategies of STH intervention in the country. Therefore, it is valuable added effort to determine the re infection rates of STHs and assessing current efficacy of chewable oral single dose (400 mg) ALB against STH infection among SC in Jimma Town, south Ethiopia.

1.2. Statement of the problem

Globally, more than 1.5 billion people or 24% of the world's population are infected with STHs. They are widely distributed in tropical and subtropical areas, with the greatest numbers occurring in SSA, the Americas, China and East Asia(20). Over 270 million PSAC and over 600 million SC live in areas where these parasites are intensively transmitted, and are in need of treatment and preventive interventions(1). There are different factors associated with increased transmission of STHs among SC in different parts of the world. Important contextual determinants for human infection are poverty, lack of sanitation and inadequate hygiene, warm climates and adequate moisture are essential for the hatching or embryonation of STHs eggs in the environment or development of larvae.

An important epidemiological feature is their highly aggregated distribution with the majority of people harboring low intensity infection, while only few individuals harbor very heavy infection(8, 21). Transmission of STHs occurs via contact with contaminated soil (*hookworm*) or consumption of egg-contaminated foods *(A. lumbricoides* and *T. trichiura*)(22). People of all ages are affected by this cycle of prevalent parasitic infection; however, SC are the most affected(21). The disease burden mainly manifested as nutritional stress and associated with poor appetite, food indigestion, mal-absorption, impaired growth and anemia. Anemia, malnutrition and STHs infections are prevalent throughout the developing nations of the world(23). Most STHs infections are asymptomatic, heavier infections can result in abdominal pain, diarrhea, malaise, weakness chronic pain and malnutrition can cause reducing school performance and productivity mainly on SC(24).

In most endemic regions of Africa, anthelmintihcs are periodically administrated to the communities, and particularly to SC, PSAC and pregnant women in order to control infection and prevent related morbidity(25). The major means of controlling STHs infection is by the periodic administering of one of the four anthelmintihcs recommended by the WHO: mebendazole (MBD), ALB, levamisole and pyrantel; while ivermectin is not recommended for the treatment of human STH(3, 19). There is a concern that continuous administration of antihelmnthics drugs may lead to drug resistant parasites (26).

Albendazole (ALB) is one of the most frequently used anthelmintics(27). A single oral dose of 400 mg ALB has been found to be effective against A. lumbricoides in terms of eggs reduction rates (EER) but less efficacious against T. trichiura and its effect on hookworm is highly variable(28, 29). Some population of STHs that are tolerant to the drug may exist. As resistance may develop rapidly when repeated mass-drug administration's (MDAs) are applied, or during mass therapy, appropriate treatment strategies should be implemented in countries endemic with STHs (30). Monitoring the change of drug efficacy is vital to institute appropriate measures timely when required.

High treatment frequency, mono-drug regimes, targeting and timing of MDA and under dosing treatments may accelerate the selection of anthelminthic drug resistant STHs. Although anthelminthic drug resistance in STHs infecting humans is not common at the moment, resistance may develop(31). Reduced efficacy of ABZ against STHs can be considered as an early warning. In addition, different generic versions of anthelminthic drugs are now available at a low cost(8, 32). As these drugs may vary in their content of active ingredient, purity, disintegration, dissolution, and bioavailability which affect their therapeutic efficacy(32), evaluating their efficacy is vital.

Determining efficacy of single dose 400mg ALB and re-infection rates of STHs could help for government and other stakeholders to design others methods to supplement the existing strategies to interrupt the re-infection rates of STHs among SC. However, there was no well documented data particularly on re-infection rates of STHs among SC in our study area.

Therefore, in order to fill these gaps the study was conducted to assess efficacy of single dose 400mg ALB on STHs after 3 weeks post treatment and re infection rates of STHs after 18 weeks among schoolchildren in Jimma Town Southwest Ethiopia.

1.3. Significance of the study

The global strategy to control STH infection is a "preventive chemotherapy" in which large-scale administration of standard anthelmintic drugs in order to reduce parasitic loads and morbidity. This control strategy is performed frequently by repeated deworming with single oral dose of 400mg ALB at a regular time interval for high-risk groups of the community; especially among SC. However, there is frequent occurrence of re-infection of STH after treating with ALB in many part of the country.

Therefore, the current study was conducted aimed to generate evidence-based information of STH infection and re infection prevalence and to assess efficacy of ALB against STH among SC in Jimma Town, southwest Ethiopia.

2. LITERATURE REVIEW

Soil-transmitted helminths (STHs) pose significant public health problem in developing countries where sanitary facilities and clean water supply are scarce. The STHs are globally distributed with several factors being associated with transmission. In such situations, an estimated 4.5 billion individuals are at risk of STHs infection and more than one billion individuals are thought to be infected, of whom 450 million suffer morbidity from their infection the majority of who are children(33). Some of the available literature on prevalence, the efficacy of 400mg ALB and re infection rate prevalence of the three STHs reviewed as follows;

Among schoolchildren observed in Asia; more than 93% of SC in Malaysia were infected by any of the STH parasites while above two-thirds of SC harbor one or more of the three STH in Africa(34). The highest prevalence of STH infection and vast majority of years lived with DALYs to STH infections occur in Asia and Africa. In SSA, there were 866 million people infected by STH as indicated by the WHO estimate 2012: the prevalence of people infected by hookworm, *A. lumbricoides*, and *T. trichiura* was 117 million (13.6%), 117 million (13.6%), and 100.8 million (11.6%), respectively(15, 35).

In Malaysia study was done among SC; the overall children were infected with one or more STHs 93.7%. The prevalence of *T. trichiura, A. lumbricoides* and hookworm infection were 84.6%, 47.6% and 3.9%, respectively. Almost half of the children had heavy *T. trichiura*; ¼ had heavy *A. lumbricoides* whereas all hookworm infections were light infection(23). In Viet Nam the prevalence of hookworm, *A. lumbricoides* and *T. trichiura* infection in the baseline were 76.2%, 19.2% and 29.1%, respectively(36). In Kenya; study was conducted to express infection intensities of STHs among SC attending seven primary schools; 44.05% were infected. *A. lumbricoides* was the most prevalent STH with 43.5%, while *T. trichiura* and hookworm infection were low with 0.82% and 0.27%, respectively(37).

In Ethiopia, the number of people living in STH endemic areas, estimated at 79 million, which comprised of 9.1 million PSC, 25.3 million, SC and 44.6 million adults. The number of individuals living in areas qualifying for STH treatment is 53.6 million, comprised of 4.6 million PASC, 17.7 million SC, and 31.3 million adults(38). Among SC; 1/3 of infected by *A.*

lumbricoides, ¼ are infected by *T. trichiura*, and hookworm infect one in eight. These make Ethiopia the second and the third high burden country in terms of *A. lumbricoides* and hookworm infection in SSA(39). The study was conducted in Ethiopia; Wondo among SC; 88.3% were found positive for one or more STH at baseline. The prevalence of any STH infection found to be 33.1% in children 3 weeks after treatment and the prevalence of hookworm, *A. lumbricoides,* and *T. trichiura* infection was 1.5%, 1.5% and 35.0% respectively(25).

A study was conducted in Jimma zone; Sigmo woreda; the overall baseline prevalence at least one of STH was 41.7%. *A. lumbricoides* was the predominant parasite 19.8% followed by *T. trichiura* 15.6% and hookworm was 1%. Out of positive for *A. lumbricoides*, 0.7% had moderate intensity infection while 19.5% had light intensity infection, out of 15.6 % positive for *T. trichiura* 9.6% had light intensity infection, while 6.6% had moderate intensity infection and 0.7% had heavy intensity of infection. The intensity of hookworm infection in all children was light(40). Similarly; in Jimma Town among PSAC, the overall prevalence STH was 46.6%. *T. trichiura* was the most prevalent 38.9% followed by *A. lumbricoides* 20.3%. Hookworm infection found in 5.5%. The majority of the infection classified as low, and only a minority classified as high(41).

In 2011; the study conducted among SC in seven different countries where STH are an endemic; the efficacy of single 400mg dose of ALB was assessed against these three STH; where the efficacy was very high for *A. lumbricoides* 99.5% and hookworm*s* 94.8% but low for *T. trichiura* 50.8%(29). In South Africa SC were treated twice with 400mg ALB and re-examined 3 weeks after the first treatment in order to assess the impact of treatment. Reinfection rates within 16 weeks after the first, 18, and 29 weeks after the second treatment. 90.0% of children infected with one or more STHs, 55.9% infected with two or more STHs and 30.9% of the children infected with all three STHs. The hookworm prevalence was 83.2%; whereas *T. trichiura* and *A. lumbricoides* prevalence were 57.2 % and 19.4%, respectively. With single dose treatment of ALB was very effective against hookworm and *A. lumbricoides* with arithmetic mean ERR of 93.2 and 97.7%, respectively. It was exceptionally ineffective against *T. trichiura with* ERR 24.8%)(42).

Recently study design was carried out in Ghana in different seasons, to assess the efficacy of single dose ALB against *T. trichiura* and hookworm in terms of ERR. After single dose ALB treatment administered to all cases at baseline assessment, later at 21 days and 3 months post-treatment. Of all the parasites found, hookworm was the most prevalent. In the dry season, the overall STH prevalence at pre- treatment was while 9% and 13% prevalence recorded at 21 days, and 3 months after treatment, respectively. However, in the rainy season, the overall STHs prevalence was 8%, while 4% and 12% recorded at 21 days and 3 months respectively after ALB treatment. Hookworm ERR of 89% in the dry season and 93% in the rainy season, while the *T. trichiura* ERR was 100% in both seasons(43).

There was systematic review and network meta-analysis carried out; to evaluate efficacies of different brands of ALB against STHs in terms ERR. All ALB showed that highest efficacy against *A. lumbricoides* and hookworm infection with ERR of hookworm infection was 89.6% and all drugs had low efficacy against *T. trichiura*(44). Similarly; in Philippines among SC revealed an ALB efficacy of 54.0% as indicated by ERR(45). Although the study were carried out in 3 countries located in three geographical areas, Haiti, Kenya and Panama among SC; the efficacy of ALB after treatment the ERR of *T. trichiura* and hookworm were 10.1% and 96.8% in Kenya respectively, while ERR of *A. lumbricoides* values indicated good drug efficacy in Panama (46). Jimma Town; the study was done to determine therapeutic efficacy of different brands of ALB against STHs, the prevalence was 45.6% and the ERR *for A. lumbricoides*, *T. trichiura* and hookworm were 97%, 99.9% and 99.9%, respectively(47). Similar to that of in the above in 2015; study was conducted in Jimma Town to assess efficacy and quality of two different brands of commonly used ALB; both brands of ALB were showed satisfactory and reduced efficacy against *A. lumbricoides* and *T. trichiura,* respectively(48).

To investigate the pattern of STH re infection rates study was conducted in 2008 in Malaysia among SC. The overall prevalence of *A. lumbricoides*, *T. trichiura* and *hookworm* infection was 65.8, 97.5 and 10.8%, respectively. After complete deworming with a 3-days course of 400 mg ALB, three months later the re-infection rate, by one or more of STHs species, was 49.5%(49). In China; similar study was carried out; the baseline prevalence of *T.trichiura*, *A. lumbricoides* and hookworm were 94.5, 93.3 and 61.3%, respectively. Most of the infected children harbored light *T. trichiura* infection 86.3%; whereas 13.1% had infection of moderate intensity. Rapid re-

infection with *A. lumbricoides* and slower re infection rate with hookworm and *T. trichiura* were observed(50). A systematic review and meta-analysis study carried out in 2012 to determine STH infection rates pre- and post-treatment; *A. lumbricoides* prevalence reached 26%, for *T. trichiura* re infection prevalence were 36% and for hookworm 30%(51).

Recently, in Tanzania; study was conducted among SC; to investigate the effectiveness of the three drug combinations against STHs in terms of ERR after 3 weeks and re infection pattern of STHs after 18 weeks post-treatment. After complete baseline and follow up, children treated with ALB-oxantel pamoate showed a higher efficacy against *T. trichiura,* ERR: 98.6 % compared to the other treatment arms. The re infection rates of *T. trichiura* 18 weeks post-treatment among all treatment arms were 37.2%. 34.6% and 25% for *A. lumbricoides* and hookworm respectively(33). In Ethiopia; Chencha district the prevalence and re-infection rates of STHs after 3 months post-treatment was 39.4% and 36.8%, respectively. The estimated prevalence of re-infection for *A.lumbricoides*, *T. trichiura*, and *hookworms* were 23.2%, 16.1%, and 41.0%, respectively. However, all infected children had light intensity of infection(52).

3. OBJECTIVES

3.1. General objective

To determine efficacy of single dose 400mg ALB and re-infection rates of soil-transmitted helminths among schoolchildren in Jimma town, southwest, Ethiopia

3.2. Specific objectives

- To assess efficacy of single dose of 400mg ALB against STHs among schoolchildren in Jimma Town.
- To assess the re-infection rates of STHs among schoolchildren in Jimma Town

4. MATERIALS AND METHODS

4.1. Study area and study period

The study was conducted from April to September 2017 in public primary schools in Jimma Town, Oromiya Region, Southwest Ethiopia. Jimma Town is located 354 km southwest of Addis Ababa. The primary schools in Jimma Town are the one implementing preventive mass chemotherapy using ALB in the control of STH infection at national level. According to the 2007, population and housing census the town has a total population of 340,400. From these, 22,419 male and 24,653 were female. Of which 27, 850 male and female attending their class in 22-governmental primary schools (Reported document 2017 from Jimma Town Education Bureau)

4.2. Study design

School based prospective cohort study design was conducted to assess efficacy of ALB and the re infection rates of STHs after three and eighteen weeks, respectively.

4.3. Population

4.3.1. Source population

All primary SC in Jimma Town in the year 2017

4.3.2. Study population

All randomly selected SC from four (Seto yido, Jimma, Hamile-19 and Dil-fire) primary schools

4.4. Sample size and sampling technique

4.4.1. Sample size determination

The required sample size was determined by using formulae; for estimating single population proportion**.**

$$n = \frac{(Z\alpha/_2)^2 \times p(1-p)}{d^2}$$

Where:

- n = sample size
- P = expected proportion of STH re infection among SC (0.368)(52)
- Z α /2 = 1.96 (at 95% confidence level)
- d = 5% marginal error

Based on the above assumptions and using single population proportion formulae; the minimum sample size of 357 required.

Finally, to minimize errors arising from the probable occurrence of non-compliance (non-responded rate), 10% of the sample size added and finally 393-SC included in the study.

4.4.2. Sampling techniques

A total of 22-public primary schools were found in the town in the study year. Four schools were selected randomly using lottery method and included in the study. The schools were Seto Yido, Jimma, Hamile-19 and Dil-fire Primary Schools. All children attending from grade one to seven and age 5-15 years at the start of the study in the four primary schools were eligible for participation. All children attending the four schools and falling into this age range invited to participate the study. In order to maintain or ensure each age class is fairly distributed among the study, in each school participants were stratified according to three age groups; (age 5–8 years, 9–12 and 13–15years). The total number of children enrolled in the study was depending on the total number of children in each school to allocate sample size.

Finally, participants from each age group were selected by systematic random sampling technique using table of random number.

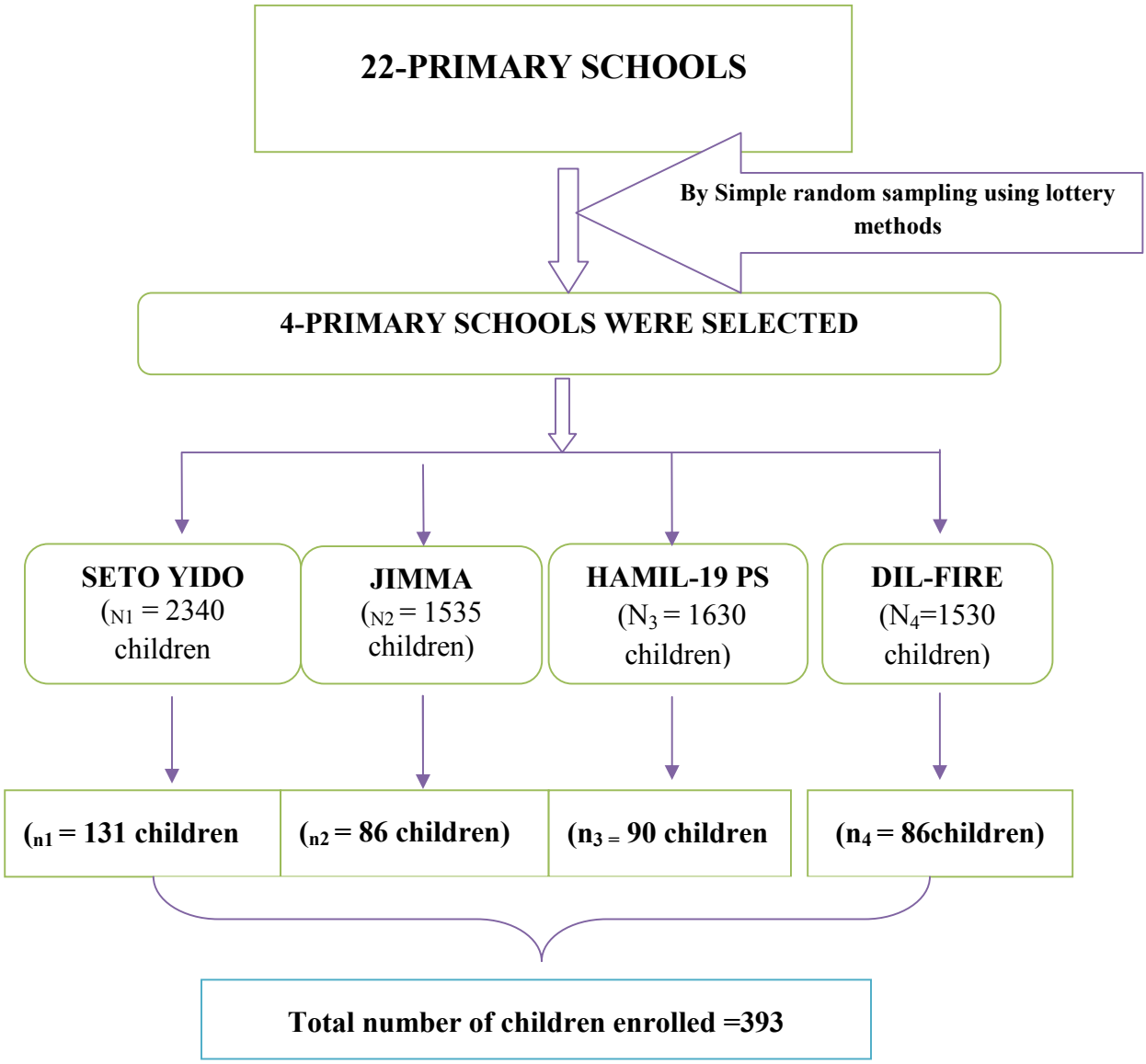

Figure 1:- Schematic presentation of sampling technique of the study.

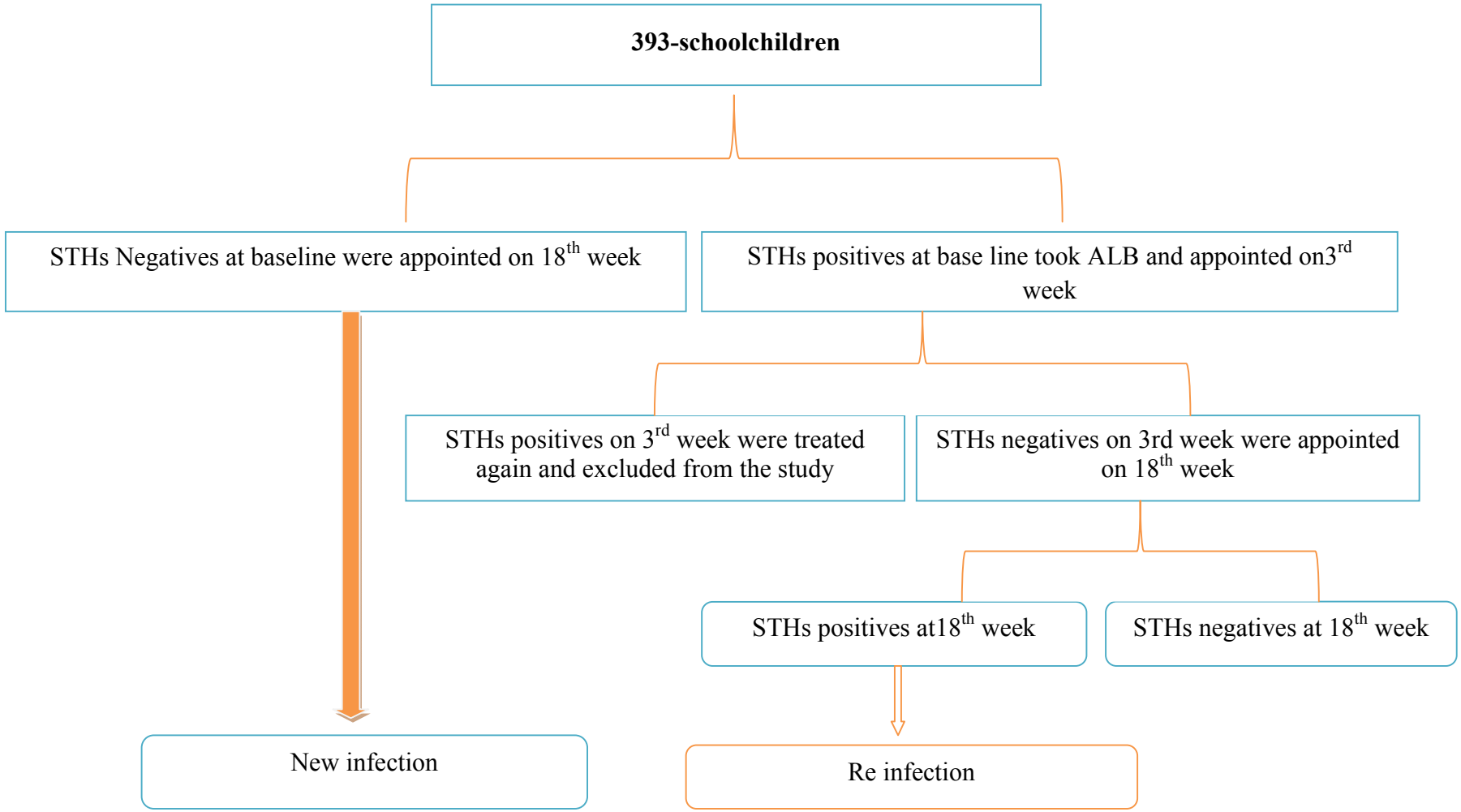

Figure 2:- Flow chart of the study design

4.5. Eligibility criteria

4.5.1. Inclusion and exclusion criteria at baseline

Inclusion criteria

- Children aged between 5 to 15years
- Children who live in the study area for at least 3 months
- Children who would have a written informed consent from their parents/guardian and also who show verbal assent
- Children who shows willing to take oral medication and participate on each visit

Exclusion criteria

- Children who have history of treatment before 3 weeks with any antibiotics and anthelmintics
- Children who vomited the administered ALB within 2 hours
- Children who unable to provide sample
- Children who not volunteer to provide sample

4.5.2. Inclusion and exclusion criteria after three weeks post-treatment

Inclusion criteria

- Positive children for any of the three STHs at baseline and taken ALB testament

Exclusion criteria

- Children who were unable to provide sample
- Children who were not volunteer to provide sample
- Children who were absent at the time of visit

4.5.3. Inclusion and exclusion criteria after 18 weeks post-treatment

Inclusion criteria

- All children who negative at baseline survey
- All children who negative result after 3 weeks post-treatment

Exclusion criteria

- Children who unable to provide sample
- Children who not volunteer to provide sample
- Children who absent at the time of visit

4.6. Study variables

4.6.1. Dependent variables

- Efficacy of single dose 400mg Albendazole
- Reinfection rate of soil-transmitted helminths

4.6.2. Independent variables

- Age
- Sex
- Baseline infection

4.7. Data collection tools

4.7.1. Questionnaire

Scocio-demographic data was collected by using semi-structured questionnaires

4.7.2. Specimen collection

About 2gm of fresh stool specimen from each study participant was collected in clean, dry and leak proof (labeled with unique ID №) stool container at base line survey, three and after eighteen weeks of treatment at each school. Collected stool specimens were immediately

transported to Jimma University Molecular Biology and NTD Laboratory Research Center for examination using Kato Katz method.

4.8. Preparation and examination of the specimen

The double Kato Katz technique was applied to process all stool samples collected from each visits. This Kato Katz diagnostic technique is recommended by the WHO to quantities STHs. The prepared fecal smear samples were examined microscopically within 30 – 60 minutes by trained laboratory professionals for the detection and quantification of STHs eggs. The actual counts of STHs eggs from both Kato Katz preparations were recorded on record format and mean of both sides were calculated. Finally, the mean count of both Kato Katz slide was multiplied by correction factors (24) to get Egg per gram (EPG).

4.9. Treatment and follow up

Based on baseline stool examination result; children who were infected with STH treated with single dose of (400mg) ALB under direct observation. Origin of 400mg/tablet ALB (Manufactured by medopharm, India, Batch №:- 5MF-41, Mfg date 06/2015 and Expire date 05/2018) was purchased from Jimma health center pharmacy in Jimma Town, Ethiopia. According to the WHO deworming guideline, after complete successful treatment requested re-sampling after 3 weeks post treatment from those only who were became positive at baseline to assesses efficacy of ALB. In this study efficacy of single-dose ALB against STHs we measured in terms ERR after three weeks post-treatment. Collected specimens were re examined for STHs egg count by using the same methods used for screening at baseline.

However, 18 weeks post-treatment stool samples were collected from all children (i.e. children negative at baseline and 3 weeks post treatment; diagnosis performed by the same procedures and re infection rates of STHs was assessed. At the end of the study all children remains infected with STH and others intestinal parasites (IPs) were treated with appropriate drugs according to the national guideline.

4.10. Data quality assurance

To ensure the quality and reliability of data, standard operating procedures (SOPs) were followed and 10% of all visits of Kato Katz slides were selected randomly and re-examined by senior Medical Laboratory technologist. (i.e. selected 1 of every 10 slides) in a blinded manner to compare with previous recordings and determine any discrepancies. Discussing for detected discrepancies with the original readers until consensus reached. Discrepancies of up to 10 % were allowed; particularly missed presence of a kind of eggs, false-positive diagnoses and egg count differences >10 eggs (counts =100 eggs) >20% (counts > 1000 eggs).

4.11. Data analysis

Statistical Package Social Sciences (SPSS) version 20 was used for descriptive data analysis like (percentage mean), frequencies range, and proportion. The results summarized and presented by tables. Calculations were also applied to assess baseline prevalence, ERR by arithmetic mean and re infection rates (Annex VI).

4.12. Ethical consideration

Ethical clearance was obtained from School of Medical Laboratory Science ethically declared by Institutional Review Board (IRB) of Institute of Health, Jimma University, with Reference № 778/09 March 2017. Permission letter written from Jimma Town education office and the ethical approval from Jimma University was provided to each school administration. Before the commencement of the study, school administration teachers and students were introduced the aim and procedures of the study. After introduction of the study, children who were voluntary to participate in the study were provided us written informed consent from their family /legal guardians.

4.13. Operational definition

- **New infection**: -Children who are negative at baseline and positive at 18^{th}week
- **Reinfection: -**Children positive at baseline who were treated successfully and negative at 3^{rd} week and became positive on 18^{th} week.

- **Schoolchildren: -**Children between the age of 5 and 15 years who are attending primary school.

5. RESULTS

5.1. General characteristics of the study participants

A total of 393 schoolchildren were participated in this study from four primary schools in Jimma Town. Of these (182/393) 46.3% were males whereas (211/393) 53.7% were females. The age of the study participants ranged from 5-15 years, with mean age of 10 years. The majority (244/393) 62.0% of the study participants were in the age group 9-12 years followed by 5-8 years(100/393) 25.5% and 13-15 years (49/393) 12.5% (Table 1).

Table 1:- Socio-demographic characteristics of schoolchildren at baseline assessment of STHs in Jimma Town primary schools, (n= 393).

Characteristics		**Frequency (n)**	**%**
Age (in years)	5-8	100	25.5
	9-12	244	62.0
	13-15	49	12.5
Sex	Male	182	46.3
	Female	211	53.7
Total		**393**	

5.2. Prevalence and infection intensities of STHs at baseline and three weeks post-treatment

The overall baseline prevalence any of the three STHs infections was (138/393) 35.1%. The highest prevalent STH was *T. trichiura* (88/393) 22.4%, followed by *A. lumbricoides,* and hookworm, which were (57/393) 14.5% and (21/393) 5.3%, respectively (Table 3). The highest prevalence of STHs found in the age group of 13-15 years (25/49) 51% and followed by aged groups 5-8 years (35/100) 35%; Even though, there is no statistically significant with age groups of the children with any of the three STHs infection. Whereas; female children is significantly associated with STHs infection (p = 0.04) (Table 2).

Table 2:- Baseline prevalence of STHs within age category and sex among schoolchildren

Variables		Total children examined	STH prevalence (%, n)	χ^2
Age (in years)	5-8	100	(35, 35/100)	2.15
	9-12	244	(32, 78/244)	
	13-15	49	(51, 25/49)	
Sex	Male	182	(35.7, 65/182)	0.04
	Female	211	(34.6, 73/211)	
Total		393	(35.1, 138/393)	

Overall; most of the study participants had light infection at baseline assessment. Out of 57/393 positive for *A. lumbricoides* 50.9 % had light, 43.9% moderate and the rest 5.2% heavy infection intensity. The only one child was positive for *A. lumbricoides* after 3 weeks post-treatment and was recorded light infection intensity. Out of 88/393 positive for *T. trichiura* more than 93% had light infection intensity, moderate and heavy infections were less frequent, after 3 weeks post-treatment about 48 (94%) children had light infection. In case of hookworm, about 95.2% had light infection, but after 3 weeks post-treatment all cases of children were recorded light infection (Table 3).

A total of 28 (20%) children were mixed infection at baseline assessment. *A.lumbricoides* with *T. trichiura*, *T. trichiura* with *hookworm* were recorded 17 (12%) and 8 (5.8%), respectively. The rest 3 (2.2%) infected with all three the STHs. About 76 (19.3%) of children were harbored others IPs other than STHs, that includes; *Schistosoma mansoni about 62 (*81.5%*), H. nana*, Taeina species and others, about 14 (18.4%) (Table 3).

5.3. Efficacy of Albendazole against soil-transmitted helminths

5.3.1. Efficacy of Albendazole against *A. lumbricoides*

In this study out of 57, 14.5% children were recorded infection with *A. lumbricoides* at baseline, majority light (50.9%) and minority moderate infection intensity (43.9%); few children were infected with heavy infection (5.2%). After three weeks post- treatment all children were negative; whereas only one child was recorded positive result with light infection. However, the

prevalence of arithmetic mean EPG of stool was 8877.6 and 12.0 before and after treatment, respectively (Table 3). Calculating the ERR according to the WHO guideline formulae; the eggs reduction rate of ALB against *A. lumbricoides* was achieved 99.9% (Table 3).

5.3.2. Efficacy of Albendazole against *T. trichiura*

At baseline; from a total of 393, 88 children were infected with *T. trichiura.* After administer ALB treatment, children provided stool samples 3 weeks post-treatment. About (51/88) 58% children were remains with *T. trichiura* infection; in contrast (37/88) 42% children were negative results. The arithmetic mean EPG was 342.1 before treatment; whereas 212.0 after taking treatment. Therefore, by using the above formulae in similar way; the eggs reduction rate of ALB against *T. trichiura* was 38% (Table 3).

5.3.3. Efficacy of Albendazole against hookworm

Out of 393, 21 (5.3%) children were infected with hookworm at baseline survey, more than 95 % was light infection intensity; mean EPG before treatment was 226.8. After swallowing ALB; (23.8%, 5/21) were became positive and their mean EPG was 105.6 and (15/21) 71.4% children were negative. However; calculating the reduced eggs of hookworm with ALB accomplished 54.4% (Table 3).

Table 3:- Prevalence, intensity of infection and arithmetic mean ERR of ALB at baseline and 3 weeks post-treatment among SC in Jimma Town, 2017.

Parameters		Soil-transmitted helminths					
		Baseline (N= 393)			After three weeks (N=136)		
		AL (%, n)	TT (%, n)	HW (%, n)	AL (%, n)	TT (%, n)	HW (%, n)
Positive children before treatment (prevalence)		(14.5, 57/393)	(22.4, 88/393)	(5.3, 21/393)	(1.8, 1/56)	(58, 51/88)	(28.6, 6/21)
Prevalence of the three STHs		35.1 (138/393)			(42.6%, 58/136)		
Negative children after 3 weeks post treatment		NA	NA	NA	(98.2, 55/56)	(42, 37/88)	(71.4, 15/21)
Arithmetic mean EPG		88977.6	342.1	226.8	12.0	212.0	105.6
Infection intensity	Light	(50.9, 29/57)	(93.2, 82/88)	(95.2, 20/21)	(100, 1/1)	(94.1, 48/51)	(100, 6/6)
	Moderate	(43.9, 25/57)	(6.8, 6/88)	(4.8, 1/21)	-	(5.9, 3/51)	-
	Heavy	(5.2, 3/57)	-	-	-	-	-
ERR of ALB		NA	NA	NA	99.9	38	54.4
Mixed infection		(20, 28/138)			-		

NA - not applicable, AL- *A.lumbricoides, TT-T.trichiura,* HW-Hookworm, n-numbers

5.4. Reinfection rates of the three soil-transmitted helminths

Out of 255 children who had negative at baseline, about 241 children from baseline and 80 children those negative after 3 weeks post-treatment were participated for re infection prevalence assessment. After 18 weeks post-treatment, the overall prevalence of three STH infections was (75/321) 23.4% (including the new and re infection). Prevalence of STHs species was (36/321) 11.2%, (46/321) 14.3%, and (10/321) 3.0% for *A. lumbricoides, T. trichiura* and hookworm, respectively (Table 6)

The re infection rates of the three STHs was (21/80) 26.3% while (54/241) 22.4% were encountered new infection prevalence. Out of these; the re-infection rate of *T. trichiura was* 27% (10/37) which is the most prevalent followed by (8/55) 14.5% of *A. lumbricoides* and (3/16) 19 % of hookworm infection (Table 6).

There were also about (72/321) 22.4% others IPs; including *S. mansoni*, about (52/72) 72.3% and (18/72) 25% was H. nana, Taeina species and others were detected. Among age between 9-12 years 12% (12/80) of children through the study were relatively more re infected than those were age between 13-15years (5/80) , 6.3% and 5-9 years 4/80) 5% ((Table 4).

Table 4:- The overall re-infection prevalence of STHs 18 weeks post-treatment by age classification among SC in Jimma Town, 2017 (n=321)

Age classification (in yrs.)	Total prevalence (%, n) (Both new and re-infection rates)	Reinfection re-infection (%, n)
5 -8	(5.9, 19/321)	**(5, 4/80)**
9-12	(14.6, 47/321)	**(12, 12/80)**
13-15	(2.8, 9/321)	**(6.3, 5/80)**
Total	(23.4, 75/321)	**(26.3, 21/80)**

Table 5:- Infection intensity of the three STHs and others IPs after 18-week post-treatment

STHs species	**Total Positive (n)**	**STHs infection intensity status after 18 weeks post treatment**					
		New infection			Re infection		
		Light (%, n)	Moderate (%, n)	Heavy (%, n)	Light (%, n)	Moderate (%, n)	Heavy (%, n)
A.lumbricoides	36	(72.2, 26/36)	(5.6 2/36)	-	(100, 8/8)	-	-
T. trichiura	46	(78.2 36/46)	-	-	(21.7, 10/46)	-	-
Hookworm	10	(70, 7/10)	-	-	(100, 3/3.)	-	-
Mixed infection	17						
A.lumbricoides and T.trichiura	13	Overall prevalence (21, 17/80)					
T. trichiura and Hookworm	3						
A.lumbricoides and Hookworm	1						
Others IPs							
S.mansoni	(72.3, 52/72)	Overall prevalence (22.4, 72/321)					
Others IPs	(25, 18/72)						

5.4.1. Reinfection rates of *A. lumbricoides*

In total, 57 of the 393 children had positive at baseline and treated; (55/56) 98.2% children became negative results 3 weeks post-treatment. After 18 weeks, (36/321) 11.2% children had acquired infection (both new and re infection children); while (8/55) 14.5% negative children were re-infected after 18 weeks post-treatment (Table 6). All re infection prevalence for *A. lumbricoides* were light (Table 5).

5.4.1. Reinfection rates of *T. trichiura*

The prevalence of *T. trichiura* at baseline was (88/393) 22.4% and positive children treated; (37/88) 42% became negative 3-week post-treatment. After 18 weeks post-treatment (46/321) 14.3% children were acquired *T. trichiura* (both new and re infection). Out of 37 participants (10/37) 27% were re- infected 18 weeks-post-treatment; whereas 36 children (36/241) 14.9% were acquired new infection (Table 6). Almost all for *T. trichiura* (both new and re infection) were light intensity (Table 5).

5.4.3. Reinfection rates of hookworm

For hookworm the prevalence at baseline was (21/393) 5.3%. Positive children were treated and (5/21) 23.8% remains positive after 3 weeks and excluded from the study. In total, 15 of 21 children (71.4%) negative and appointed them 18 weeks post-treatment. Out of them 10 children (10/321) 3.0% had acquired infection (both new and re infection), whereas (3/16) 19% were re infected after 18 weeks post-treatment with hookworm (Table 6). All children (re and new infection) were infected with light infection (Table 5).

Table 6:- Baseline infection and re infection prevalence of the three STHs among SC in Jimma Town.

STH species	% prevalence at baseline (%, n)	Infection status of STH after 18 weeks post-treatment (%, n)		
		Total positive prevalence (%, n)	New infection prevalence (%, n)	Reinfection rate (%, n)
A. Lumbricoides	(14.5, 57/393)	(11.2, 36/321)	8.7% (28/241)	(14.5, 8/55)
T. trichiura	(22.4, 88/393)	(14.3, 46/321)	(14.9, 36/241)	(27, 10/37)
Hookworm	(5.3, 21/393)	(3.0, 10/321)	(2.9, 7/241)	(19, 3/16)
Total	(35.1, 138/393)	(23.4, 75/321)	(22.4, 54/241)	(26.3, 21/80)

6. DISCUSSION

Soil-transmitted helminths (STH) have been the most prevalent infection among peoples living in resource-limited developing countries. In recent, the control of STHs has got significant attention by many governments and stakeholders(53). Frequent preventive chemotherapy control program has been on progress by ALB treatment of choice against infection with all three STHs in most Ethiopian primary schools to reduce the morbidity. The aim of present study was to assess the efficacy of ALB in terms of ERR and re infection rates of STHs in Jimma Town among primary SC which provides appropriate information about the re infection prevalence of STHs in community.

From the total of 393-SC, 35.1% were infected by any of the three STHs; most infection was *T. trichiura* 22.4%, followed by *A. lumbricoides* 14.5% and 5.3% hookworm.

The overall prevalence any of the three of STHs infection at baseline in our study (35.1%) is comparable with the others previous finding study conducted among SC; in Ghana 29% (43), Kenya 44.05%(37), South Ethiopia, Wondo Genet 33.1%(25), Chencha 39.4%(52), Jimma Zone and Jimma Town, which was reported 41.7% and 46.6% (40, 41) respectively. In contrast, the 35.1% prevalence of STHs infection in SC that we reported in Jimma Town was lower than the 90% prevalence reported in South Africa(42) and 53.5% prevalence reported in 2013 Jimma Town(54).The predominant type of STH at baseline was *T. trichiura* 22.4% similar study conducted in Jimma Zone, Sigmo Woreda 15.6% (40), 23.1% in Jimma Town(55) and lower than the study done among PSAC in Jimma Town 38.9% (41). The prevalence of *A. lumbricoides* in present study was (14.5%) similar with study conducted in Amhara region 14%, (56) and 23.6 % in Jimma(55). The observed difference might be arisen from age variation between the studies and baseline prevalence of STH infection.

This study confirm the reduced efficacy of single dose (400mg) ALB against STHs in terms of ERR, particularly for *T. trichiura* and hookworm infection among SC in Jimma Town. The ERR for *A. lumbricoides, T. trichiura* and hookworm were 99.9%, 38% and 54.4%, respectively.

Our finding indicated that, ALB efficacy in terms of ERR was satisfactory for *A. lumbricoides* 99.9% was comparable with recommended thresholds of ALB efficacy categorized by the WHO

in terms of ERR, (ERR > 95% = Satisfactory (19, 57). There is also agreement with study conducted in seven different countries (Brazil, Cambodia Cameroon, Ethiopia, India, Tanzania and Vietnam) 99.5% (29), Haiti 99.9% and Kenya 97.3% (46). Philippines 93% (45), Jimma Town 97% (47), South Africa 97.7% (42), Jimma Town with different brands of ALB, (i.e. Bendex; 98.7% and Ovis 97.8% (48). Eggs reduction rate of *A. lumbricoides* in our study showed also higher than study conducted in Panama was done with different diagnostic techniques 60% and 89.9%(46), the most probable differences were due to methods used for diagnosis, sample size variation or might be due to difference brands of ALB was used.

In our study ERR of *T. trichiura* was similar to results reported from South Africa 24.8% (42), Haiti 49.7%, Kenya 10.1% (46), Philippines 54% (45) and 50% in seven different countries (Brazil, Cambodia Cameroon, Ethiopia, India, Tanzania and Vietnam)(29). In Jimma Town two different brands of ALB (i.e. 24.4% for Bendex and 20.4% for Ovis(48). Comparing the present finding with other studies higher finding was also observed; Like study carried out in Jimma Town and Tanzania 99.9% and 98.6%(33, 47), respectively. The study carried out in 6 different countries where STHs are an endemic the ERR of *T. trichiura* was 64.5% (58); which indicated twice higher than the present study. In Panama also ERR was 99.5% and 65.1% by using Kato Katz and FLOTAC technique, respectively (46). The most probable difference indicates that effectiveness of combination of drugs and variation in laboratory diagnostic methods were used.

The EER of ALB for hookworm (54.4%) in the present study is similar with a study done in Panama ERR was 54% (46). But higher results were obtained study conducted in Jimma Town 99.9%(47), study conducted in seven different countries 94.8%(29), 96.2% in 6 different countries (Brazil, Cambodia Cameroon, Ethiopia, Tanzania and Vietnam) (58), South-Africa 93.2% (42), Ghana and 89% on dry and 93% rainy season, respectively (43), in Jimma Town using different brand of ALB (i.e. Bendex ERR: 88.7% and Ovis ERR: 98.1%) (48) which is also higher comparing with the present result. These differences were might be due to effectiveness difference between different brands of ALB or time difference during assessing impact of ALB or way of eggs counting technique or might be due to baseline prevalence of STHs infection in the study area.

The overall one or more STHs re infection rate in the present study was 26.3% in line with study conducted in Ethiopia, Chencha district 36.8% (52) and lower than Malaysia 49.5% and 79.6% after 3 and 6 months deworming, respectively (49). This difference might be due to sample size and time variation

The re infection rate of *A. lumbricoides in* our study 14.5%, which is agreement the re-infection rate study conducted in Ethiopia, Chencha 23.2%. But lower than re-infection rate in Pemba Island, Tanzania 34.6% (33). Unlike the above; higher of re infection rate with *A. lumbricoides* in rural Yunnan, China 75.8 % and 83.8% after 4 and 6 months treatment, respectively(50), in rural KwaZulu-Natal/South-Africa was 97.7% (42). The most probable reason for lower re infection rate in our present study due to regular MDA implementing in community resulted in low baseline infection which causes lower re-infection rate prevalence and also longer follow up.

The re-infection rate of *T. trichiura* (27%) in our study is in agreement with study done rural KwaZulu-Natal/South-Africa 24.0% (42). Another study were conducted in Pemba Island, Tanzania 37.2% (33) and Chencha 16.1% (52) slightly similar comparing with the present study. The re infection rate of hookworm in our study 19% which also slightly in agreement with finding in China and Tanzania 30 % and 25% (33, 50), respectively. But higher re infection rates Chencha was 41.0% (52), South Africa 93.2% (42). The main reason for this difference might be due to difference in the baseline prevalence of hookworm infection and impact of MDA in our study area as well, South Africa is separate ecological zone from our study area.

7. CONCLUSION

The present study showed that after effective treatment of SC with single dose 400mg ALB there is reduced efficacy of ALB particularly for *T. trichiura* and hookworm detected and there is re-infection rates of the three STHs were observed. Even if, ALB works well against *A. lumbricoides* it is the best suitable tool for the control of STHs infections. Yearly deworming might achieve decreased morbidity.

8. RECOMMENDATION

However, we recommend other complimentary strategies should be in place like health education, sanitation and improvement both in personal and environmental hygiene to minimize the re-infection prevalence. Furthermore, long-term investigation is an urgent needed in order to develop an appropriate preventive method or strengthening the existing strategy to sustain the effect of deworming as well as to control re infection of STHs.

9. LIMITATION OF THE STUDY

The limitation of this study;

- We doesn't assess the socio-economic status of the study participants
- Inability involving all age groups population
- Some children were absent and transfer to another school during assessing re-infection

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ANNEXES

Annex I:- Information sheet

1. English version

Name of organization

Jimma University, Institute of health, School of Medical laboratory sciences

Topic

"Assessment of efficacy of ALB and re infection rates of STHs among SC in Jimma Town" Southwest, Ethiopia 2017

Investigator

Mr.Alemu Worku

Advisors

1. Mr.Daniel Dana
2. Mr. Mio Ayana

Purpose of the research

The purpose of this research will be carried out to assess efficacy of ALB and re-infection rates of STHs among SC in Jimma Town

Voluntary of the study participants

The right of study participants to be enrolled or to refuse respected. All study subjects were participating freely after being informed about the study purpose. They will have the right to refuse answering questions, providing stool and withdraw from the study at any time. We will not expect from you to give a reason to stop the study.

The risks and benefits

The research will be carried out with minimum or no risk for the subjects. There will be no discomfort during sample collection. We would fully respect the individual, the culture and the society. Moreover, all children excreting STH eggs offered an appropriate treatment from us, in line with the national guidelines free of charge.

Confidentiality

Ensured by making laboratory results record format anonymous and by publishing research findings in a way that were not relate to the study subject

Contacts

If you have any question, suggestion comments or anything that is not clear**;** please contact us;

1. Mr.Alemu Worku Cell phone +251- 912213080; email alemuworku11@gmail.com
2. Mr.Daniel Dana Cell phone +251- 917800188, email; danidana2000@gmail.com
3. Mr. Mio Ayana Cell phone +251 917805479;email,mioayana@yahoo.com

Finally, we would like to say thank you for taking time to hear the information given and willing to participate the study.
If you are clear with the information provided and agree to participate, please sign the next page on the consent form.

Annex II: - Consent form

Assessment of efficacy of ALB and re infection rates of STHs among SC in Jimma Town" Southwest, Ethiopia 2017

1. English version

I, the undersigned individual, am oriented about the objectives of the study. I have informed that all of my information will be kept confidential and used solely for this study. In addition, I have been well informed that my name will not be asked and unique identification is not required. If I want to withdraw from the study anytime along the process, I will not be obliged to continue or give reasons for doing so. However, my agreement to participate in this study is with the assumption that, the information and the specimen that I provide will help greatly to the assessment of efficacy of ALB against STH and re infection rates of STHs.

Signature;_____ Date; _____

ጅማ ዩኒቨርስቲ ጤና ሳይንስ ኢንስቲቲዩት ፣ላቦራቶሪ ሳይንስ ት/ት ክፍል

2. ስለጥናቱ ኢንፎርሜሽን በአማርኛ ግልባጭ (Amharic version)

<u>ድርጅት</u>

ጅማ ዩኒቨርስቲ ፤ ጤና ሳይንስ ኢንስቲቲዩት፤ ላቦራቶሪ ሳይንስ ት/ት ክፍል

የጥናቱ ርዕስ

“በአፈር አማካይነት የሚተላለፉ የሆድ ትላትሎችን በአልቢዳዞል መድኃኒት ከታከሙ ከ3 ሳምንታት በኃላ ስለመድኃኒ ቱፍቱንነት ለማወቅ እና ከ18 ሳምንታት በኃላ ደግሞ ትላትሎቹ ስለመመለሳቸዉ እና አለመመለሳቸዉን ለመገምገም በጅማ ከተማ የመጀመሪያ ደረጃ ት/ቤት ዉስጥ የሚማሩ ተማሪዎች ላይ የሚደረግ ምርምር ነዉ።።

ተመራማሪዎች

1. አቶ አለሙ ወርቁ (ተመራማሪ)
2. አቶ ዳንኤል ዳና (አማካሪ)
3. አቶ ሚአ አያና (አማካሪ)

መብት

አንተ/ቺ በዚህ ጥናት ለመሳተፍ በሎቶሪ ዘዴ ወይንም በቀላሉ አልፎ አልፎ በተማሪዎች ብዛትና በሚፈለገው የተማሪዎች ብዛት መሰረት የተመረጥ/ሽ ሲሆን፤ ነገር ግን በዚህ ጥናት በመሳተፍህ/ሽን ለየት ያለ የምታገኝዉ/ኚዉ መብት የለም ።። ነገር ግን የምታገኘው/ የምታገኚዉ ጥቅማ ጥቅም ካለ በማንኛውም መንገድ የማይገድብብህ/ሽ ነው።።

ጥቅም

የምታገኘው/ የምታገኚዉ ጥቅማትቅሞች ባይኖሩም በምርመራ ወቅት በሰገራህ/ሽ ውስጥ ትላትሎቹ ከተገኙ በነፃ መድሀኒት የማግኘት መብት አለህ/ሽ ።።

ሚስጢራዊት

ሁሉም ስለጤንነትህ/ሽ የሚጠቅሱ የምርመራ ውጤቶች ሚስጢራዊነታቸው በሚገባ የሚጠበቅነው ።። በማንኛውም ሪፖርት ወይም ህትመት አትታወቅም/ቂም።።

የሚያደርሰዉ ጉዳት- ይህን በማድረግህ/ሽ ምንም አይነት ጉዳት የሚደርስብህ /ሽ የለም፡፡

ለተለያዩ ጥያቄዎ ችአድራሻ

1. አቶ አለሙ ወርቁ ስልክ ቁጥር +251- 912213080; email alemuworku11@gmail.com
2. አቶ ዳንአልዳና ስልክ ቁጥር+251- 917800188, email; danidana2000@gmail.com
3. አቶ ሚአ አያና ስልክ ቁጥር+251- 917805479;email, mioayana@yahoo.comመጠቀም ትችላላችሁ፡፡

በጥናቱ ለመሳተፍ ፍቃደኛ ከሆንክ/ሽ በሚቀጥለዉ ገፅ ላይፍርምልኝ ፡፡ አመሰግናለሁ

ጅማ ዩኒቨርስቲ ጤና ሳይንስ ኢንስቲቲዩት ፣ላቦራቶሪ ሳይንስ ት/ት ክፍል

2. የስምምነት ማረጋገጫ (Amharic version)

ማብራሪያ

ይህ የስምምነት ቅጽ የተዘጋጀዉ በጅማ ከተማ የመጀመሪያ ደረጃ ት/ቤት የሚማሩ ተማሪዎች ላይ በአፈር አማካይነት የሚተላለፉ የሆድ ትላትሎችን በመድኃኒት ከታከሙከ 3 ሳምንታት በኃላ ስለመድኃኒቱፍቱንነት ለማወቅ እና ከ18 ሳምንታት በኃላ ደግሞ ትላትሎቹ ስለመመለሳቸዉ እና አለመመለሳቸወን ለማወቅ ለሚደረገዉ ምርምር የተማሪዎችን ፍቃደኝነት ለመግለፅ የተዘጋጀ የስምምነት ማረጋገጫ ቅጽ ነዉ።።

እኔ ስሜ ሳይጠቀስ በመለያ ኮድ ብቻ የምለየዉ ተማሪ ስለምርምሩ በቂ ገለጻ ከተደረገልኝ በኃላ በሆዴ ዉስጥ ለሚገኙትን ትላትሎች ለማወቅ ምርመራ እንዳደርግ (ሰገራ እንድሰጥ) ተጠይቂያለሁ።። እነዚህ ትላትሎች እኔ በምሰጠው ናሙና ውስጥ የሚገኙ ከሆነ አስፈላጊው ህክምና የሚሰጠኝ እንደሆነ፣ ናሙናውን በመስጠት ምንም አይነት ጉዳት የማይመጣብኝ መሆኑን፤ የምርመራው ውጤት በሚስጥር እንደሚጠበቅ ተገልፆልኛል።።ከሁሉም በላይ በጥናቱ ላይ መሳተፍ በፍቃደኝነት ላይ የተመሠረተ መሆኑና ለመሳተፍ ፍቃደኛ ከመሆኔ በፊት እንዳስብበት በቂ ጊዜ ተሰጥቶኛል ።።

ስለዚህ በጥናቱ ለመሳተፍ የወሰንኩት ስለሁኔታው በሚገባ ከተረዳሁ በኋላ በጥናቱ ሂደት ውስጥ በፍቃደኝነት ለመሳተፍ ተስማምቻለሁ ።።

ፊርማ _____ ቀን _____

3. Garagalcha afaan oromo (Afan Oromo version)

Maqaa dhaabbataa

Universitii Jimma, Dhaabbata Fayyaa, Mana Barumsa Saayinsii Laaboratorii

Mata dureee qorannoo

Raammolee dhukkuba deddebisanii namatti fidan barbaadufi human albeendazolin ammam akka isaan balleessuu danda'u ijoolle barumsa sadarkaa 1ffaa baratan irratti qorachuu

Maqaa qorataa

1. Obbo.Alamuu Workuu
2. Obboo.Daani'el Daanaa
3. Obboo. Mi'oo Ayyaanaa

Seensa

Ati qorannoo kana irratti akka hirmmattu affeeramteeta. Kanaafuu atis haaluma kanaan sababa qorannoon kun adeemsifamuuf sirritti erga hubatte booddee gaaffii siif dhiyyatu deebisuun akka nu gargaartu kabajaan isin gaaffanna

Faayidaa qorannoo

Qorannoon mataa duree raammolee dhukkuba deddebisanii namatti fidan barbaadufi human albeendazolin 400mg ammam akka isaan balleessuu danda'u ijoolle barumsa sadarkaa 1ffaa baratan irratti qorachuu jedhu irraatti adeemsifamu kun faayida guddaa kan qabu yoo ta'u kunis namoota raammoon kun keessatti argame qorichi kafaltii malee kennamaf. Akkasumas haalli fayyuumma keessan akka mirkaneeffatnif isin gargaara

Mirga isin qabdan

Isin qorannoo kana keessatti akka hirmmatan kan akka carraatin baratoota jiran waan filamtanif itti hirmaachu yookin dhiisuuf mirga guuttuu qabdu. Kanaaf ofii keessani murteessitanii akka itti hirmaattani kabajaan isin gaafacha yoo qabdani jidduun keessa ba'u barbaaddan ni dandeessu.

Faayidaa

Qorannoo kanatti hirmaachu keessanif faayidaa baayyee argatuu. Kanneen keessa inni duraa waa'ee fayyuumma keesani raammolee kanniin irra bilisa ta'uu keessan nimirkaneeffattu. Akkasumas yoo rakkoon jiraata kaffaltii malee qoricha argattu

Icciitii

Mirgi sagalee keessan bilisan kennuufi iccitin keessan sirritti eegama. Tarii dhoksaatti sagalee keessan laachuu yoo barbaaddan mirga guuttuu qabaachuu keessan isini mirkanneessa odeeffannoon isin irraa argamu lakkii dhoksaa (koodii) waan funaanamuf odeeffanno isin laattan eenyulee adda baasee beekuu hin danda'u.

Teessoo

Yoo gaaffii qabaattan amma bilisaa tatani nagaafachuu dandeessu.

1. Obbo.Alamuu Workuu Lak. bilbila +251- 912213080., email alemuworku11@gmail.com
2. Obboo.Daani'el Daanaa Lak. bilbila +251- 917800188, email danidana2000@gmail.com
3. Obboo. Mi'oo Ayyaanaa Lak. bilbila +251- 917805479, email, mioayana@yahoo.com
   ni dendessu Galattommaa.

Annex III: - Study participants registration check list

Participant registration check list for assessing efficacy of ALB and re infection rates of STHs among SC in Jimma Town" Southwest, Ethiopia 2017

Name of school _____Date _____

S.№	L.code	Full Name of Children	Age	Sex	Grade and section	Family phone №	Remark

The format is filed by:_____ Date _____

The format approved by: _____Date _____

Annex IV:- Standard Operating Procedures (SOPs)

Diagnosis of STH by double Kato Katz thick smear techniques for assessing the efficacy of ALB and re infection rates of STHs among SC in Jimma Town" Southwest, Ethiopia 2017

1. Kato Katz thick smear technique

Principle: In the Kato Katz thick technique faces are processed through a mesh screen to remove larger particles. A portion of the sieved samples were then transferred to the hole of a template on a slide, After filling the hole, the template was removed and the reaming sample (approx. 41.7gm) was covered with a piece of cellophane socked in a glycerol – malachite green solution (glycerin). The glycerol "clears" the fecal materials from around the eggs and the malachite green stains, the background of the field. The eggs counted and the number calculated per grams (gm) of stool.

To collect stool specimen from SC for diagnosis of STH by Kato Katz thick smear techniques; for the assessment of the efficacy of ALB and re infection rates of STH among SC in Jimma Town the following Materials and reagents were needed

2. Materials and reagents

2.1. For collecting stool sample

- Plastic containers with wooden applicator sticks
- Permanent marker, pen and pencil
- Toilet paper or absorbent tissue and investigation form

2.2. For specimen preparation and examination

- Kato set
- Microscope slides
- Forceps
- Graduated volumetric apparatus
- Disposable gloves
- Flat- bottom jar with lid
- Newspaper or glazed tile
- Microscopes

- Tally counter
- Scissors
- Timer
- Slide rack
- Specimen

2.3. For cleaning material;

- Bleach (3 or 5% Bleach)
- Heavy-duty rubber gloves
- Bucket, Powder soaps
- Brush

2.4. For treatment 400mg ALB

3. Preparation of Kato Katz solution for working

Glycerol-malachite green or Glycerol-Methylene blue solution (1 ml of 3% aqueous malachite green or 3% Methylene blue was added to 100 ml of glycerol and 100 ml of distilled water and mixed well). The solution was poured onto the cellophane strips in a jar and left for at least 24 h prior to use.

NB:- Templates of different sizes have been produced in different countries. However, we use a hole of 6 mm on a 1.5 mm thick template, 41.7 mg and cellophane as cover slide (soaked in Glycerol – malachite green solution before 24 hrs)

4. Sample collection

- Explain why stool needed and how to deposit stool sample over clean newspaper
- Distribute labeled container and materials for stool collection to bring an appropriate volume approximately 6gm, fresh and own stool samples

5. Sample rejection criteria

- Liquid or diarrheal samples at first sampling
- Insufficient sample
- Unlabeled or unclear ID № or if any uncertainty sample was found

6. Procedure for Kato Katz thick smears preparation

6.1. Place a small mound of faucal material on newspaper and press the small screen on top so that some of the feces sieved through the screen and accumulate on top. Scrape the flat-sided spatula across the upper surface of the screen to collect the sieved feces.

6.2. Place template with hole on the center of a microscope slide and add stool from the spatula so that the hole was completely filled.

6.3. Using the side of the spatula pass over the template to remove excess stool from the edge of the hole

6.4. Remove the template carefully so that the cylinder of feces is left on the slide.

6.5. Cover the stool material with the pre-soaked cellophane strip.

The strip must be very wet if the stool is dry and less so if the stool is soft (if excess glycerol solution is present on upper surface of cellophane wipe with toilet paper). In dry climate excess glycerol will retard but not prevent drying.

6.6. Invert the microscope slide and firmly press the stool sample against the hydrophilic cellophane strip on another microscope slide. The fecal material is spread evenly between the microscope slide and the cellophane strip. It will be possible to read newspaper print through the smear after clarification

6.7. Carefully remove slide by gently sliding it sideways to avoid separating the cellophane strip or lifting it off. Place the slide on the bench with the cellophane upwards. Water evaporates while glycerol clears the stool.

6.8. For all except hookworm eggs, keep slide for one or more hrs at ambient temperature to clear the fecal material prior to examination under the microscope. To speed up clearing and examination; slide can be placed in 40°C incubator or kept in direct sunlight for several minutes

6.9. *Ascaris* and *Trichuris* eggs will remain visible and recognizable for many months in these preparations. Hookworm eggs clear rapidly and will no longer be visible after 30-60 minutes.

7. Reading and reporting the results

The smear was examined the whole slide in a systematic manner and the number of eggs of each species reported. Later multiply by the appropriate number to give the number of EPG of stool (by 24 for a 41.7 mg template)

Annex V:- Laboratory results reporting and Quality control format

1. Laboratory result registration form

Laboratory result registration form for diagnosis of STH by double Kato Katz thick smear technique for assessing efficacy of ALB and re infection rates of STHs among SC in Jimma Town" Southwest, Ethiopia 2017

Name of school _____Date _____

L.code	Slides	*A. Lumbricoides*	*T. trichiura*	Hookworm	Others IP	Initial
	A					
	B					
	A					
	B					
	A					
	B					
	A					
	B					
	A					
	B					
	A					
	B					
	A					
	B					

The format is filed by:_____ Date _____

The format approved by: _____Date _____

2. Quality control chart for diagnosis of soil-transmitted helminths

Quality control form for diagnosis of STH by double Kato Katz thick smear technique for assessing efficacy of ALB and re infection rates of STHs among SC in Jimma Town" Southwest, Ethiopia 2017

Part. code	Slide	Original reading (OR)						Quality control reading										Acceptance status		
		KK — AL	KK -- TT	KK -- HW	KK -- SM	KK -- others	Initial OR	KK -- AL	KK -- TT	KK -- HW	KK -- SM	KK -- Others	Initial OR	KK -- AL	KK -- TT	KK -- HW	KK -- SM	Accepted	Corrected	Remarks
	A																			
	B																			
	A																			
	B																			
	A																			
	B																			
	A																			
	B																			
	A																			
	B																			
	A																			
	B																			

Annex VI:-Formula for assessing baseline, re infection rates of STHs and ERR with thresholds of albendazole efficacy

In helminthology, the baseline prevalence of the STHs; the following formula was used

$$\text{Prevalence} = \frac{\text{Number of children testing positive}}{\text{Total number of children screened}} \times 100\%$$

The Arithmetic mean number of EPG of feces commonly used as indicator for estimating the severity of infection in a population and the changes that occur after treatment (for assessing the efficacy of ALB) measured in terms of ERR, using the formula, the WHO recommended guideline(17, 19).

$$\text{ERR} = \frac{\text{Arithmetic mean EPG before Rx. - Arithmetic mean EPG after Rx.}}{\text{Arithmetic mean EPG before Rx}} \times 100\%$$

From the two samples (pre and post-treatment) and calculate ERR; Evaluation was classified and interpreted into different level by comparing the observed ERR. Satisfactory when ERR is superior or equal to the reference value; Doubtful when ERR is inferior than the reference value by less than 10% points, Reduced when ERR is inferior to the reference value by at least 10% points. The criteria applied for this classification by the WHO (57) (Table7).

Table 7:- Thresholds of ALB efficacy against the three common STH infection

№	Criteria	STHs		
		A. lumbricoides	*T. trichiura*	Hookworm
1	Satisfactory	ERR > 95%	ERR > 50%	ERR > 90%
2	Doubtful	95% > ERR > 85%	50% > ERR >40%	90% > ERR >80%
3	Reduced	ERR < 85%	ERR < 40%	ERR < 80%

Re-infection was calculated by the following formula considering children who successfully treated at base line and became negative 3 weeks post-treatment and who presented with infection at the end of 18 weeks post-treatment.

$$\text{Reinfection rates} = \frac{\text{Total children became re-infected after 18 weeks post-treatment}}{\text{Negative children after 3 weeks post-treatment}} \times 100\%$$

Annex VIII: - Determination of infection intensity of soil-transmitted helminths

Intensity of infection was estimated indirectly by counting the mean number of eggs per gram of feces (EPG) and categorized using thresholds recommended by the WHO. Accordingly, it is indicated, for *A. lumbricoides*, Light infection (1-4,999 EPG), Moderate infection (5000 - 49,999EPG) and Heavy infection (above 50,000 EPG). For *T.trichiura* Light infection (1-999 EPG), Moderate infection (1000-9,999 EPG) and Heavy infection above (10,000 EPG) and For Hookworm, Light infection (1-1,999 EPG Moderate infection (2000-3,999 EPG) and Heavy infection (above 4000 EPG)(59, 60).

Annex IX:- Declaration

I hereby declare that the work embodied in this research thesis carried out by me under direct supervision of the advisor(s), School of Medical Laboratory Sciences, Faculty of Health Sciences, Institute of Health, and Jimma University. This work has not been submitted in part or full in any University or Institution for any Degree or Diploma. I further endorse that this work is the property of Jimma University and all rights in this regard are reserved with Jimma University.

Name of student:_____ Signature: _____Date:_____/_____/_____

This thesis been submitted for examination with approval of my advisors:

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2. Name of advisor: _____Signature: _____Date:_____/_____/_____

Name of internal assessor: _____

Signature: _____Date:_____/_____/_____

Name of external assessor:_____

Signature: _____Date:_____/_____/_____

APPROVAL SHEET OF THESIS

As a member of the board of examiners of the master of science thesis open defense examination, I certify that I have read, evaluate the thesis prepared by Alemu Worku , and examined the candidates as well. I recommended that the thesis be accepted by fulfilling the thesis requirements for the degree of Master of Science in Medical laboratory.

Internal Examiner_____

Signature: _____Date:_____/_____/_____

External Examiner_____

Signature: _____Date:_____/_____/_____