

Prevalence of Intestinal Parasites Co-infection and Associated Factors Among Pulmonary Tuberculosis Patients in Adama Town, East Shoa, Oromia, Ethiopia

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Abstract: Tuberculosis and parasitic infections are co-endemic in many parts of the world. The treatments of many patients with tuberculosis are under the influence of Intestinal parasite infection that worsens the progression of this disease. Hence, adequate information on co-infection rate is needed to undertake the integrated prevention and control program. Therefore this study aimed to assess the prevalence of intestinal parasite co-infection and associated factors among pulmonary tuberculosis patients in Adama Town, East shoa, Oromia, Ethiopia. A facility based cross sectional study was conducted from November 2018 up to January 2019 using multi stage random sampling technique. Data on socio-demographic characteristics and potential risk factors for intestinal parasite co-infection was collected by standardized semi structured interviewer administered questionnaire. One gram stool samples for direct saline microscopy, formol-ether concentration technique and modified acid fast staining was collected and processed in accordance to the standard parasitological techniques and procedures at Adama Science and Technology University Higher Health Center Laboratory. Data entry was done by EPI info version 7, and analyzed using SPSS version 22.0. Pearson chi-square and multiple logistic regression models were used to identify associated factors. Overall co-infection rate of intestinal parasites was 75 (21.4%) (95% CI: 17.1-25.4). Protozoans' account 40 (11.4%) and Helminthes were 35 (10.0%). The predominate isolates were Entamoeba histolytica 30 (40%) and Ascaris lumbricoides 12 (16.0%) from protozoans and helminthes, respectively. Eating raw vegetables/ fruits without washing or peeling [AOR]: 5.311 (2.089-13.506), habit of not using soup [AOR]: 11.238 (3.134-40.296) and Body mass index < 18.5 kg/m² [AOR]: 15.337 (5.860-40.142) were significant determinants of intestinal parasite among pulmonary tuberculosis patient. In general overall co-infection rate of intestinal parasites in this study was high (21.4%). Therefore, integrating screening and mass deworming of parasite into existing tuberculosis program should be considered.

Keywords: Intestinal Parasite Co-infection, Pulmonary Tuberculosis, Adama, Ethiopia

1. Introduction

Parasitic infections and tuberculosis (TB) are both major public health problems worldwide affecting over 3.5 billion people and 9.6 million new cases respectively [1, 2]. In

addition, helminth infections and TB are both common in low and middle income countries and share a great degree of geographical overlap, making co-infection common [3]. Moreover, Tuberculosis and intestinal parasites affect primarily low social and economic level populations, living clustered in precarious habitation settings [4]. Similar to TB,

it has been estimated that one third of the global human population is infected with intestinal parasites [5].

Helminths are reported as the most common parasites found in TB patients and TB is also most prevalent in communities where people are helminthes infected [6]. As result soil-transmitted helminth infections have been evaluated as epidemiological risk factors for developing active TB [7]. While helminth and MTB infection mechanisms are vastly different with MTB being a single celled organism and helminthes being multicellular, several authors have demonstrated the negative association between helminthes and mycobacterial infections [8, 9]. The probable link for parasite interaction with MTB infection rests on the divergent immune response [9, 10]. In addition, parasitic infections affect the nutritional status of the individual leading to immunological alterations that promote a decrease in the efficacy of the immune response, favoring the occurrence of other bacterial infections like TB [10].

The high prevalence of intestinal parasites indicated an increased morbidity in TB patients [11]. Moreover, epidemiological studies of intestinal parasite co infection among patients with TB have been conducted in many developing countries where TB and intestinal parasite were prevalent. In Brazil, it was found that 19.6% of patients with TB from the community had intestinal parasite [10]. A study in China suggested that 14.9% of patients PTB from hospitals were infected with parasite [12]. In India the prevalence accounts 32% of TB patient had intestinal helminth infection [13], hospital based study in Gonder founds 70.9% pulmonary TB had one or more parasite [14]. These findings indicate that Intestinal parasite among patients with is common in marginalized, low-income and resource-constrained regions around the world and the diagnosis and treatment of many patients with TB are under the influence of intestinal parasitic infection.

The problem associated with co-infection emanates from mutually antagonistic responses induced by the extracellular helminthes and those induced by the intracellular Mtb, that can result in impaired (or cross-regulated) host responses to either of the infecting pathogens [1]. Helminthes infections typically direct the immune system toward type 2 responses, characterized by high levels of the cytokines IL-4 and IL-10, which can antagonize IFN γ production and its biological effects [15]. In general different studies shows TB patients with helminthes infections present with more severe pulmonary disease, diminished anti-Mycobacterium tuberculosis immunity, and diminished responses to anti-TB chemotherapy [7, 16].

Most of the common parasites species are concurrent with MTB increase antibacterial therapy intolerance and deteriorate prognosis of disease [13]. Moreover several authors' indicate co-infection results in impaired immune Responses to TB [17–20] and it can also resulted poor clinical response to TB therapy [21–23]. Therefore, it is worthwhile to explore co-infection with TB and intestinal parasites because co-infection increases the complexity of

control and prevention of TB and parasitic diseases in co-endemic areas [13].

Building on previous studies parasitic co-infections negatively influence the outcome of TB treatment and favor persistent TB infection, however; screening and deworming of TB patient for parasite is not integrated in routine management of TB in most health care system in Ethiopia. This raises important research questions about the public health implications of parasitic infection with MTB in terms of pathogenesis and treatment outcomes of this disease. However, very limited data is available in Ethiopia that is generated from Gondar, Northwest Ethiopia [14, 15, 24] and one study southern Ethiopia [25]. As to the best of our knowledge, no similar study has been conducted in Oromia and study area so far.

By taking the above into consideration, this study is designed to collect data on the prevalence of intestinal parasite co infection and associated factors among TB patients attending treatment in selected health facilities in Adama town, so that control and prevention of parasite infection be a part of the solution to a pandemic TB.

2. Method and Materials

2.1. Study Area and Setting

The study was conducted in Adama town located to the east of Addis Ababa at the distance of 99 km. According to the census report in 2007 conducted by the Central Statistical Agency of Ethiopia (CSA), the town has a total population of 341,796. Adama town has 1 government teaching hospital, 7 government health centers, 4 private hospitals, 94 private clinics with different level of which 17 special private clinics, 4 Non-Governmental clinics and 108 drug store and Pharmacies. Currently they are 20 health facilities serve as tuberculosis treatment centers in adama town of which 8 governmental, 8 private and 4 nongovernmental. Among 8 governmental health facilities serve as tuberculosis treatment center, 5 were randomly selected and namely; Adama Hospital Medical College (AHMC), Geda health center, Adama Health Center, Biftu health center and Boku Shenan Health center.

2.2. Study Design and Period

A cross sectional study was conducted from November 2018 to January 2019.

2.3. Population

2.3.1. Source Population

All tuberculosis patients that were on tuberculosis treatment and active PTB patient in Adama Town.

2.3.2. Study Population

Randomly Selected pulmonary tuberculosis patient who were visited TB clinic at Adama Town selected health facilities for during the study period.

2.4. Inclusion and Exclusion Criteria

2.4.1. Inclusion Criteria

The study includes pulmonary TB patients who present themselves to TB clinic for treatment and active PTB patient during the study period.

2.4.2. Exclusion Criteria

Patients who had taken anti parasitic drug within three months before data collection, severely ill patients who were unable to respond to research questions.

2.5. Sample Size Determination and Sampling Procedures

2.5.1. Sample Size Determination

Sample size was determined by using single population proportion formula by taking the prevalence for co infection rate of intestinal parasites and pulmonary TB from one of the study in among tuberculosis patients in Arba Minch town in 2017, which was 26.3 [25]. Using the following assumptions

where=(Absolute level of precision) or margin of error (d^2) of 4% at 95% confidence level when prevalence ranges between (10%-90%) [26] With non-response rate of 10%. Total sample size calculated for first objective was 344, whereas the sample size for second objectives becomes 370. The biggest sample size obtained out of all calculations was taken as optimum sample were 370 used to answer all study objectives.

Sample Size Determination for Objective one

$$n = \frac{(z/\alpha_2)^2 \times P(1-P)}{d^2}$$

$$n = \frac{(1.96)^2 \times 0.263(1-0.263)}{(0.04)^2} = 465$$

This the calculated sample size 465 is <10,000 of source population (954), and n/N is >0.05 a finite population correction is used to minimize the sample size:

$$n = \frac{N * n}{N + n}, n = \frac{954 * 465}{954 + 465} = 313$$

Table 1. Sample Size calculation for Objective two for associated factor for Intestinal parasite Co-Infection among Tuberculosis patient by epi info StatCalc.

Variables	Power	OR	CI	Ratio unexposed/ exposed	%outcome unexposed	Sample size
Raised poultry or livestock	80	3.96	95%	0.55	35.48	89
Ever labored barefoot	80	4.54	95%	1.93	65.88	126
Residence	80	3.175	95%	3.53	27.7	162
Habit of washing vegetables	80	2.208	95%	2.203	18.4	336

Source [12].

2.5.2. Sampling Technique

A multi stage random sampling technique was used to select the study participants. First from 20 public health facilities serve as tuberculosis treatment center in Private, Governmental and Nongovernmental; Governmental Treatment Center was randomly selected by simple random sampling. Second from Eight (8) governmental health facilities serve as TB treatment centers in Adama town: Adama hospital and seven health centers: Adama hospital and four health centers (Adama health center, Geda health center, Boku Shenan health center and Biftu health center) was selected with simple random sampling technique. The data obtained from those selected health facility in the last consecutive six months shows totally 954 eligible pulmonary tuberculosis patients were visited TB clinic of those facilities. Accordingly by dividing 954 pulmonary tuberculosis patient to our sample size 370 (954/370) we obtained sampling interval k of 3. From 1-3 pulmonary tuberculosis patient, picking a patient on random for the initial start, then pulmonary tuberculosis patients was selected systematically by adding 3 on initial number by using TB registration book as a sampling frame.

2.6. Variables of the Study

2.6.1. Dependent Variables

The dependent variable of the study was Intestinal parasite co-infection among PTB patients.

2.6.2. Independent Variables

The independent variables of the study were:

Socio demographic factors like Age, Sex, Occupation, Educational level. Residence.

Behavioral factor and environmental factor like hand washing habit, raw meat eating habit, swimming habit, shoe wearing habit washing vegetable habit, raise cattle, use of night soil, finger trimming, soap usage water source, have and use latrine, food source.

Medical factor (health related) like BCG vaccination status, Deworming history, Previous TB exposure, HIV status, Body mass index, History of diarrhea, Duration of TB treatment, TB treatment category.

2.7. Operational Definitions

Intestinal parasite: Intestinal parasites are organisms that live in the gastrointestinal tract of humans [27].

Intestinal protozoa: Do a diverse group of unicellular single celled parasites inhabit the intestinal tract of high vertebrate hosts including humans [19].

Intestinal Helminthes; The term “helminthes” refers collectively to wormlike parasites and encompasses two phyla of major human parasites: Platy helminthes (flatworms), which include tapeworms and flukes and Nematoda (roundworms) [28].

Body Mass Index (BMI); WHO universal classification of BMI values through a set of cut-off points to classify the weight conditions: <18.5 Underweight; 18.5 - 25.0 Normal weight; ≥25.0 Overweight [29].

2.8. Data Collection Procedures and Data Quality Assurance

Data on socio-demographic characteristics and potential risk factors for intestinal parasite was collected by semi structured interviewer administered questionnaire. The questionnaire adopted after reviewing different literature of similar studies and it modified and adapted based on the situation of the study area to collect information on intestinal parasite infection and associated factors from tuberculosis patient in Adama town tuberculosis centers. It was prepared in English, and translated in to Amharic and afaan Oromo language by native speakers and experts of the languages and re-translated back to English and Afaan Oromo language to check for any inconsistencies in the meaning of words and concepts. The final version of the questionnaire was used for data collection. A single fresh fecal sample of 1 gm. was collected in a clean wide mouth container and intestinal parasites was identified using direct wet mount, formol ether concentration and modified Ziehl–Neelsen staining as described in the SOP [27]. Direct wet mount microscopy was performed by smearing a well-mixed and small amount of fecal sample (~2 mg) on a glass slide with normal saline solution and cover slide by transporting sample immediate laboratory within 15 min. Then the slide was examined with light microscope using 10X and 40X objectives. Actively motile protozoan parasites together with other helminthes were identified based on motility and morphological structure. Differentiation of *Entamoeba histolytica* and *Entamoeba dispar* was made by identification of engulfed red blood cells and by staining the nuclei. The remaining fecal sample was subjected to the formol ether concentration technique. Briefly, a portion of fecal sample was mixed with 10% formalin solution and sieved through double layered gauze. The filtrate was transferred to a 15ml volume plastic test tube and formalin was added to reach the volume of 12ml and 3ml of diethyl ether was added. Then the tube was shook vigorously and centrifuged for 5 minutes at 3000 rpm. The sediments were observed under light microscope using 10X and 40X objectives. A portion of the pellet was smeared on a glass slide and stained with Ziehl–Neelsen staining methods to look for *Cryptosporidium species C.cayatensis* and *Isospora belli*. The stained slides was examined under microscope using 100X oil immersion [12]. Negative results were reported after assessing the whole smear under 10 x objective. Investigators supervised in all aspects of data collection and laboratory procedures.

The validity of the study Results was assured by applying and following quality control measures during the total process of the laboratory procedures. All materials, equipment, and procedures were adequately controlled. Negative and positive control slides was used to check the functionality of microscope as well as the accuracy of laboratory professional engaged in conducting the study. Every 10th specimen is reexamined for quality control and a discrepancy of 10% or less for the whole study is regarded acceptable. The initial examination was performed by a senior laboratory technician experienced in microscopic identification of intestinal parasites while the quality control examination was performed by an experienced

another experienced laboratory technologist without the knowledge of the results from the initial examination. The quality of data was controlled starting from the time of questionnaire preparation data collectors and their supervisor was trained for two days. The questionnaire and Checklist was examined by experts to the area of study for content validity. Pretests of 5% were performed on 19 pulmonary tuberculosis patients to check the reliability and validity of the questionnaires in Hawas health center before study. The questionnaire was prepared in English, and translated into Amharic and Afaan Oromo language experts of the languages and re-translated back to English and Afaan Oromo language to check for any inconsistencies in the meaning of words and concepts. The final version of the questionnaire was used for data collection.

2.9. Data Processing and Analysis

Data entry was done by EPI info version 7 and exported to SPSS version 20.0 for analysis. Basic assumptions like data normality for continuous variables was done by histogram with curve and Kolmogorov-Smirnov test was used whose *P*-value ($p < 0.05$) corresponding to rejection of normality. In the bivariate analysis, the Pearson chi square tests was used to identify associations between participants ‘characteristics and intestinal parasite co infection by computing crude odds ratios (ORs) with 95% CIs. For variables with $p \leq 0.25$ in the bivariate Analysis, Multiple logistic regression model was subsequently employed with (adjusted ORs with 95% CIs) of those risk factors that were found to be statistically significant by the bivariate analysis. Association between variables were considered statistically significant only if a two-sided *P*-value < 0.05 at 95% confidence level. Multi-Collinearity test was carried out to see the correlation between each explanatory variable, using Variance Inflation Factor (VIF) and tolerance test the values for both tests would be within the normal cut-off points i.e. $VIF > 5$ or $tolerance < 0.2$ considered problematic to the model. The Hosmer Lemeshow goodness of fit test was used to evaluate the overall fit of the final models generated.

2.10. Ethical Considerations

The study was ethically approved by Institutional Review Board (IRB) of Adama Hospital Medical College. The letters of approval and cooperation from the Oromia Regional Health Beouro was addressed to concerned departments and to the selected health institutions. Written informed consent was obtained from all study participants and in case of children Under 18, assent was obtained from the respective guardians. Once the consent was obtained from all study subjects stool sample was collected. Those who would not want to participate in the study would not be obliged. Information that was obtained in the course of investigation was kept confidential. At the completion of the study and in accordance with the local treatment policies, anti-protozoa treatment and anti-helminthic treatment was offered to all the participants found to be infected with intestinal protozoa and/or helminths by communicating with physician, TB focal person and directors of each health facility.

3. Results

3.1. Socio-demographic and Behavioral Characteristics of Study Participants

A total of 350 PTB patients were included in this study with response rate of 94.6%. One hundred twenty one of the study participant was from Geda health center while 73, 66, 54 and 36 were recruited from AHMC, Adama Health Center, Biftu health center and Boku Shenan Health center respectively. Out of 350

participants 202 (57.7%) of the respondents were males. The median age of study participants were 29 years with IQR 24-38. Majority 313 (90%) of the study participants live in urban area (Table 2). Majority 321 (91.7%) of the study participants commonly cook their food at home. Among those having frequent habit of eating raw vegetables/fruits, 201 (57.4%) wash or peel off before consumption. Most of study participants 327 (93.4%) and 279 (79.7%) wash their hand before meal and after defecation respectively (Table 3).

Table 2. Socio demographic factors of PTB patient in Adama town selected health facilities from November 2018-january 2019.

VARIABLES		FREQUENCY	
		Number (n)	Percent (%)
Sex	Female	148	42.3
	Male	202	57.7
Age category	≤18	14	4.0
	19-45	290	82.9
	46-65	34	9.7
	>66	12	3.4
Marital status	Single	144	41.1
	Married	206	58.9
Educational status	Elementary and above	254	72.6
	Illiterate	96	27.4
Residence	Urban	313	89.9
	Rural	35	10.1
Average monthly income	>100 dollars (2800birr)	52	14.9
	≤100 dollars	298	85.1

Table 3. Behavioral factors of PTB patient in Adama Town selected health facilities from November 2018-January 2019.

VARIABLES		FREQUENCY	
		Number (n)	Percent (%)
Common food source	Home	321	91.7
	Hotel	29	8.3
Washing/peeling of raw vegetables before use	Often or always	201	57.4
	Never or occasionally	149	42.6
Habit of eating raw meat	Never or occasionally	292	83.4
	Often or always	58	16.6
Hand washing habit before meal	Yes	327	93.4
	No	23	6.6
Hand washing after defecation	Soap and water	279	79.7
	Water only	71	20.3
Frequency of hand washing habit	Often or always	258	73.7
	Never or occasionally	92	26.3
Soup usage	Yes	288	82.3
	No	62	17.7
Have and use latrine	Yes	324	92.6
	No	26	7.4
Ever walked bare foot	No	149	42.6
	Yes	201	57.4
Have you farmed before	No	316	90.3
	Yes	34	9.7
Raised cattle	No	246	70.3
	Yes	104	29.7
Raised pet	No	184	52.6
	Yes	166	47.4
Use of night soil for farming	No	308	88.0
	Yes	42	12.0
Finger trimming habit	Trimmed	245	70.0
	Untrimmed	105	30.0
Water source for drinking	Pipe	330	94.3
	river/lake/stream	20	5.7
Swimming habit	No	182	52.0
	Yes	168	48.0

3.2. Medical/Health Related Characteristics of Study Participants

Out of 350 participants 161 (46%) and 133 (38.0%) have history of diarrhea and gastro intestinal discomfort within 3 month before data collection respectively. Majority 331 (94.6%) have no history of deworming within a year. All of

them were tested for HIV of whom 46 (13.1%) were sero positive. Out of 350 around 122 (34.9%) them had BMI<18.5 kg/m². During the time of interview, 293 (83.7%) of the participants have already started anti-TB treatment while the rest 57 (16.3%) were contacted as soon as diagnosed before the start of treatment (Table 4).

Table 4. Health related factors and anthropometric measurement of PTB patient attending selected health facilities in Adama Town from November 2018-january 2019.

VARIABLES		FREQUENCY	
		No	%
History of diarrhea within 3 month	No	189	54.0
	Yes	161	46.0
Other GIT discomfort within 3 month	No	217	62.0
	Yes	133	38.0
Deworming history	No	331	94.6
	Yes	19	5.4
BCG vaccination status	No	132	37.7
	Yes	218	62.3
Previous TB exposure	No	199	56.9
	Yes	151	43.1
Type of pulmonary TB	Negative for AFB	237	67.7
	Positive for AFB	113	32.3
Started TB treatment	No	57	16.3
	Yes	293	83.7
Course of treatment	First	295	84.3
	Retreatment	55	15.7
	<2	217	62.0
Duration of treatment	2-4	99	28.3
	>4	34	9.7
	>18.5kg/m	228	65.1
Body mass index	<=18.5kg/m	122	34.9
	Negative	304	86.9
HIV status	Positive	46	13.1

Keys: GIT; Gastro Intestinal Discomfort HIV: Human Immuno Deficiency Syndrome.

3.3. Prevalence of Intestinal Parasites co Infection Among PTB

Among 350 PTB patients, overall co-infection rate of intestinal parasites was 75 (21.4%) (95% CI: 17.1-25.4). The prevalence of intestinal protozoans' account 40 (11.4%) and that of intestinal helminthes were 35 (10.0%). Among intestinal protozoans' *Entamoeba histolytica* 30 (40%) predominant parasite isolated from PTB followed by *Giardia lamblia* 6 (8%). Totally six helminthes were detected among which *Ascaris lumbricoides* was with the highest frequency 12 (16%) followed by *Tania spp* 6 (8%) (Figure 1).

3.4. Factors Associated with Intestinal Parasite co-infection

From the result of bivariate and multiple logistic regression model it was observed that being female in sex, not utilizing Soup For hand washing, never washing/peeling of raw vegetables/fruit, Not hand washing habit before meal, using water only for Hand washing after defecation, Not Having and using latrine, Ever walked with bare foot, History of diarrhea within 3 month, Finger trimming habit, Other GIT discomfort, Body Mass Index and HIV status had significant association with parasite co-infection (p-

value<0.25): However, after fitting those variables into multiple logistic regression model, only 5 of them had significant association (P<0.05). Those include rarely/occasionally washing/peeling of raw vegetables/fruit before consumption [AOR]: 5.311 (2.089-13.506), Hand washing by water only after defecation [AOR]: 2.780 (1.016-7.609), Habit of not using soup [AOR]: 11.238 (3.134-40.296), those that complain GIT discomfort [AOR]: 7.175 (2.250-22.879) and Body Mass Index<18.5 kg/m² [AOR]: 15.337 (5.860-40.142) (Table 5).

In a multivariate logistic regression model, we found that PTB patient that eat raw vegetables/ fruits without washing or peeling 5 times more likely to be infected by intestinal parasite compared to PTB patient that often or always wash/peel raw vegetables (adjusted odds ratio [AOR]: 5.311 (2.089-13.506). Those PTB patient that wash their hand by water only after defecation, the odd of being infected by parasite increased by 3 fold compared to PTB patient that wash their hand by soup and water (adjusted odds ratio [AOR]: 2.780 (1.016-7.609) (Table 5).

The odd off having parasitic co-infection increased by 11 fold among pulmonary tuberculosis patients that had not habit of soup using as compared to those that did [AOR]:

11.238 (3.134-40.296). Furthermore, our study finding indicated that the odd off having parasitic co-infection 7 times more likely higher among pulmonary tuberculosis patient that complain GIT discomfort as compared to those did not [AOR]: 7.175 (2.250-22.879). Lastly our study

finding revealed that the odd off having parasitic co-infection increased by 15 fold among pulmonary tuberculosis patient whose body mass index<18.5 kg/m² as compared to its counter parts [AOR]: 15.337 (5.860-40.142).

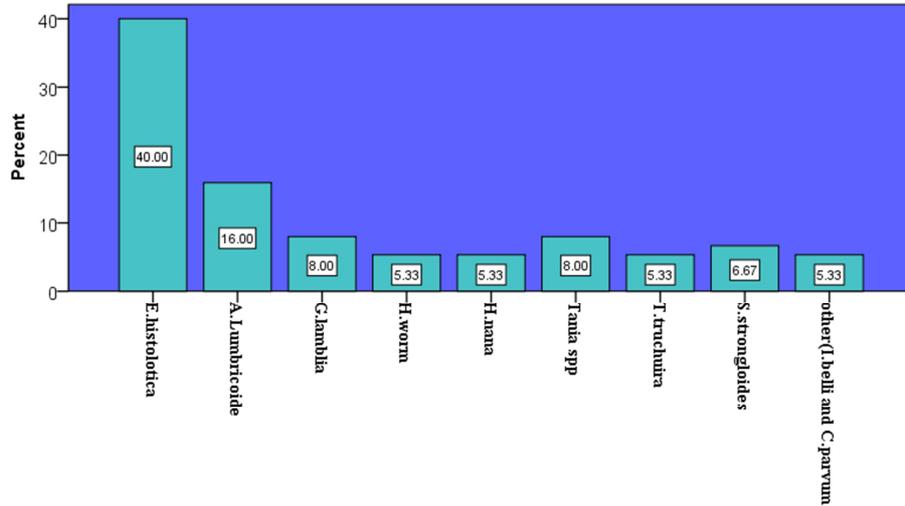


Figure 1. Prevalence of intestinal parasite among pulmonary TB patient attending selected treatment centers in Adama Town from November 2018-january 2019.

Table 5. Bivariate chi-square and multiple logistic regression analysis for associated factors with intestinal parasite among PTB patients attending selected health facilities in Adama Town from November 2018-january 2019.

Variables	Category	Parasite co-infection		Crude OR (95% CI)	AOR (95% CI)
		Present	Absent		
Sex	Male	23 (15.5%)	125 (84.5%)	1.00	
	Female	52 (25.7%)	150 (74.3)	1.88 (1.1-3.3)*	
Education status	primary& above	56 (22.0%)	189 (77.9%)	1.00	
	Illiterate	19 (19.8%)	77 (80.2)	.87 (.487-1.563)	
Residence	Urban	63 (20.1%)	250 (79.9)	1.00	
	Rural	12 (31.4%)	23 (65.7)	1.819 (.846-3.91)	
Soup usage	Yes	35 (12.2%)	253 (87.8%)	1.00	1.00
	No	40 (64.5%)	22 (35.5%)	13.1 (7.0-24.6)*	11.2 (3.1-40.3)
Washing raw vegetables/fruit	Often /always	17 (8.5%)	184 (91.5)	1.00	1.00
	Never/occasionally	58 (38.9%)	91 (61.1)	6.899 (3.8-12.5)*	5.3 (2.01-13.5)
Habit of eating raw meat	Often /always	64 (21.9%)	228 (78.1%)	1.00	
	Never/occasionally	11 (19.0%)	47 (81%)	.834 (.409-1.700)	
Hand washing before meal	Yes	64 (19.6%)	263 (80.4%)	1.00	
	No	11 (47.8%)	12 (52.2%)	3.8 (1.59-8.9)*	
Hand washing after defecation	Soap& water	40 (14.3%)	239 (85.7%)	1.00	1.00
	Water only	35 (49.3%)	36 (50.7%)	5.8 (3.275-10.3)*	2.780 (1.02-7.61)
Have and use latrine	Yes	62 (19.1%)	262 (80.9)	1.00	
	No	13 (50.0%)	13 (50%)	4.2 (1.8-9.56)**	
Ever walked bare foot	Yes	20 (13.4%)	129 (86.6%)	1.00	
	No	55 (27.4%)	146 (72.6%)	2.4 (1.4-4.27)**	
Finger trimming habit	Trimmed	38 (15.5%)	107 (84.5%)	1.00	
	Untrimmed	37 (35.2%)	68 (64.8)	1.53 (1.8-2.8)**	
History of diarrhea within 3month	No	16 (8.5%)	173 (91.5%)	1.00	
	Yes	59 (36.6%)	102 (63.4)	6.1 (3.5-11.44)**	
Other GIT discomfort	No	20 (9.2%)	197 (90.8%)	1.00	1.00
	Yes	55 (41.4%)	78 (58.6%)	6.9 (3.9-12.3)**	7.2 (2.2-22.88)
BMI	>18.5kg/m	21 (9.2%)	207 (90.8%)	1.00	1.00
	<=18.5kg/m	54 (44.3%)	68 (55.7%)	7.8 (4.4-13.8)**	15.3 (5.8-40.1)
HIV status	Negative	57 (18.8%)	247 (81.2%)		
	Positive	18 (39.1%)	28 (60.9%)		

Key; BMI: Body Mass Index, *: significant at bivariate, PTB: Pulmonary Tuberculosis, GIT: Gastro Intestinal Discomfort, HIV: Human Immuno Deficiency Virus, OR: Odd ratio.

4. Discussion

The current study finding indicated that the overall prevalence of intestinal parasite among PTB patients was found to be 21.4%. This finding is higher than a study conducted in different areas, namely Brazil 19.6% [10], China 14.9% [30]. This observed difference might be due level of awareness about intestinal parasite transmission and prevention and economic status of the population might be essential determining factor for the decreased prevalence of intestinal parasite in Brazil and China as compared to the current study area. This finding is lower compared to previous studies conducted elsewhere in Ethiopia: three studies from Northwest Ethiopia 70.9% [14], 40.5% [31], 40% [15], Woldia General Hospital in North Wollo 49% [32], Arba Minch hospital 26.3% [25] and Addis Ababa 22% [33]. The existence of such variations may be explained by the differences in practices of personal hygiene, study period, environmental sanitation, health promotion practices, geographical location, periodic mass deworming made by urban health extension worker and type of diagnostic sensitivity.

In current study, we observed that an overall rate of helminthes infection accounts 10% among patients with PTB. This finding is in line with study done in Brazil 10.1% [10], higher than that of study done in china 7.6% [30], much lower than study done in southern India 32% [13], Tanzania 22.9% [34], North West Ethiopia by 70.9% [14] and Arba Minch hospital 24.4% [25]. Compared with other studies, findings of these studies give a clue that infection spectrum of intestinal helminthes might have gradually changed over recent years this probably because of this majority of study participants comes from urban they have better hygienic practices, beside that they are accustomed to wear shoes and not to defecate in the open which may explain the low prevalence of intestinal helminthes infection. In addition the laboratory protocol followed may justify for those differences. In both previous studies, 3 stool samples were collected and examined from each participant before ruling out intestinal helminthiasis. However, in the present study, we have collected and examined a single stool specimen for the sake of time and logistic problems.

In current study, we found that overall rate of intestinal protozoan infections accounts 11.4%. This finding is higher than study done in Brazil 9.8% [10], China 7.9% [30]. This probably due to elevated environmental contamination, food and water contamination and poor personal hygienic behavior by those affected. Furthermore, most of raw vegetables/fruits consumed by those parasitized PTB patient comes from unknown source that may highly contaminated by intestinal protozoans.

The proportion of helminthes species prevalent among PTB patients varies according to a few other studies. This study identifies *A. lumbricoides* was the predominant helminthes parasite with rates of 3.4%, the finding of this study was in agreement with study done by Alemu and Mama

in Arba Minch hospital in which *A. lumbricoides* was the most prevalent parasite with rates of 11.3% [25] and Aynalem Alemu in Addis Ababa [33], un like this study, study done in Tanzania shows that *S. stercoralis* was the most prevalent parasite with rates of 17.0% [35], More over *Hook worms* were the most frequent helminthes in China, Egypt and Northwest Ethiopia with rates of 4.3%, 16.5% and 11.1% respectively [11, 30, 36]. This difference probably due to Climate conditions, socio-demographic characteristics and living standards of the population are believed to determine the type of helminthes existing in various TB endemic areas.

The most frequently isolated intestinal protozoan in this study was *E. histolytica/dispar* with rate of 8.6%. The finding was in line with study done in Brazil 3.0% [10], But, it was in disagreement with a study done in china in which *Blastocystis hominis* (6.2%) [30] and Arba Minch Hospital in which *G.Lamblia* 4.7% [25] predominant protozoan parasite isolated from pulmonary tuberculosis patient respectively. This study shows variations in the frequency and type of parasites might be due to differences Low personal hygiene practices Such as not washing hands after defecation, failure to use soap during hand washing as it independently associated with parasitic infection. The finding in agreement with people who never used soap for washing hands were at 30 times higher risk of parasitic infection [37].

Among many possible associated factors, we found that PTB patient that eat raw vegetables/ fruits without washing or peeling 5 times more likely to be infected by intestinal parasite compared to PTB patient that often or always wash/peel raw vegetables. Alemu and Mama and Xin-Xu Lia etal also report similar finding [12, 25]. The possible justification for this association could be fruit and vegetables consumed by majority of urban community from comes unknown source in that contaminated vegetables and fruit may act as vehicle parasitic infection. In this study, we found that those PTB patient that wash their hand by water only after defecation, the odd of being infected by parasite increased by 3 fold compared to PTB patient that wash their hand by soup and water. Moreover, PTB patient that could not use soap 11 times more likely to be infected by intestinal parasite compared to counter parts. This finding is supported by practicing hand-washing with soap after defecation, a 63.6% to 78% reduction in load of intestinal parasite achieved without chemotherapy [38]. This probably explained by water alone did not remove microbes from our hand but soap has capability to carry germs from hand.

We found that BMI<18.5 was independently associated with intestinal parasite, PTB patient whose BMI<18.5 the odd being infected by parasite increased by 15 fold compared to those with normal weight. This finding is consistent with Alemu and Mama [25], Alemayehu etal [24] and Aynalem Alemu [33]. This generally indicate Parasitic infections affect the nutritional status of the individual leading to immunological alterations that promote a decrease in the efficacy of the immune response, favoring the occurrence of other bacterial infections [10]. lastly we noticed PTB patient

that complain gastro intestinal discomfort 7 times more likely to be infected by parasitic infection compared to those who had not complain gastro intestinal discomfort. A study done in Tanzania and Gonder Northern Ethiopia shows that, TB-HIV co infected patient less likely to be affected by parasite compared to TB only [15, 34]. Our finding does not support this finding.

5. Strength and Limitations of Study

5.1. Strength of Study

This multicenter study was conducted among five (5) public health facility of Adama town and would have a better representation of the study participants and generalizability of the result. Probability multistage random sampling method were used that can have better representation of study population.

5.2. Limitations of Study

Prevalence of intestinal parasite was determined by examination of single stool specimen from each study participant. Thus, we could not access the intra- and interstool variation of egg output. Being cross sectional nature of study it did not show cause-effect relationships between variables.

6. Conclusion and Recommendation

6.1. Conclusion

Over all co-infection rate of intestinal parasite among PTB patients in this study was high (21.4%). The prevalence of intestinal protozoans' account 11.4% and that of intestinal helminths were (10.0%). Among intestinal protozoans' *Entamoeba histolytica* predominant parasite isolated from PTB and among helminths *Ascaris lumbricoides* was with the highest frequency. Never/ occasionally Washing/peeling of raw vegetables/fruit, Hand washing after defecation, failure to use soap, GIT discomfort and Body Mass Index < 18.5 independently associated with Intestinal parasite co-infection among PTB patient.

6.2. Recommendation

1. We recommend pulmonary tuberculosis patient to protect their personal hygiene by regular hand by washing before meal and after defecation, to use soap regularly and to avoid consumption of uncooked vegetables.
2. We recommend health care provider in particular that work on tuberculosis program to provide health education on good hygiene habits as prevention of intestinal parasite among patients with PTB may lead to a positive tuberculosis outcome. Moreover, screening of TB patient for parasite also seeks great attention from health care provider.
3. We invite other researcher to undergo experimental

studies to accurately put impact parasite on treatment outcome of TB.

4. We recommend policy makers to integrate intestinal parasite screening and mass deworming of pulmonary tuberculosis patient into existing tuberculosis program to enhance holistic management of both parasite and pandemic tuberculosis simultaneously.

Acronym/Abbreviation

AHMC: Adama Hospital Medical College
 BCG: Bacille de Calmette et Guerin
 HIV: Human Immuno deficiency Virus
 IFN γ : Interferon Gamma
 LTBI: latent Tuberculosis Infection
 MTB: Mycobacterium tuberculosis
 NTDs: Neglected Tropical Diseases
 PTB: Pulmonary Tuberculosis
 SOPs: Standard Operational Procedures
 STH: Soil-Transmitted Helminthiasis
 TGF: Tumor necrosis Growth Factor

Compliance with Ethical Standards

Conflict of Interest

The authors declare that they have no competing interests.

Financial Disclosure

All the expense for this original study was covered by principal investigator.

Ethics Approval

Ethical Approval was obtained from Oromia Region Health Bureau, Adama Town Health Office and Adama Hospital Medical College.

Key Message

Original data is available on request.

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