Prevalence of Intestinal Parasitic Infection and Associated Risk Factors among Food Handlers in Wachemo University Students' Cafeteria, Southern, Ethiopia

By

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MSc Thesis Submitted to Jimma University, Institute of Health, Faculty of Public Health, Department of Environmental Health Sciences and Technology in partial fulfillment of the requirements for the degree of Master of Science in Environmental Health

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ABSTRACT

Background: In developing countries like Ethiopia intestinal parasites is one of the food-borne diseases. Therefore, regular evaluation of food handlers for their health status, including screening for the prevalence of parasites is important.

Objective: To assess prevalence and predictors of intestinal parasitic infections among food handlers working in Wachemo University students' cafeteria, Southern, Ethiopia.

Methods: A cross- sectional study design was conducted. Socio-demographic data and associated risk factors were gathered using semi structured questionnaire. The prevalence of parasites was assessed following standard parasitological method assisted by professionals. Data were coded, cleaned and entered into Epi data 3.3.1 and then analyzed using SPSS Version 20. Statistical significance was assessed using Binary and Multiple logistic regression model, odds ratios and 95% CIs, P values less than 0.05 was used as cut off point for statistical significance. Informed consent was obtained and confidentiality assured.

Results: A total of 212 food-handlers working at cafeteria of Wachemo University were participated in the study. The majority of study participants were females 135(63.7 %). Of the 212 stool specimens examined, about 63 (29.7 %) were found to be positive for different intestinal parasites with the most prevalent parasite *Ascaris lumbercoids* 27 (12.7%) followed by Hookworms 14(6.6%), *Giardia lamblia* 10(4.7%), *Taenia saginata 5* (2.4%), and Mixed infection 7 (3.3%). Out of the 212 finger nail content specimens, 12 (5.6%) were positive for one or more parasites. The prevalence of intestinal parasites was significantly associated with risk factors such as sex, education, finger nail status and hand washing with soap and water after toilet use.

Conclusion: The present study revealed a moderate prevalence of intestinal parasites among food handlers. Since most of the intestinal parasites are transmitted by the feco-oral route, food handlers could be an important source of infection to the students and general population. So that giving health education about personal hygiene for food handlers recommended.

Key words: Intestinal parasites, Prevalence, Food handlers

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LIST OF ABBREVIATIONS

AOR	Adjusted Odd Ratio
CI	Confidence Interval
COR	Crude Odd Ratio
ETB	Ethiopian Birr
FDREMOH	Federal Democratic Republic of Ethiopia Ministry of Health
IPI	Intestinal Parasitic Infection
NTD	Neglected Tropical Disease
OR	Odd Ratio
SOP	Standard Operation Procedure
SNNPR	Southern Nations, Nationalities, and Peoples' Region
SPSS	Statistical Package for Social Science
STH	Soil Transmitted Helminthes
WHO	World Health Organization
WU	Wachemo University

1. INTRODUCTION

1.1 Background

Study conducted in different countries showed that intestinal parasitosis was public health problems worldwide. As a result of food borne microbial pathogens in developed world 30% of the population suffers from diarrheal disease. Because of food borne disease about 2 million death occurred annually in developing country (Mama and Alemu 2016). Reported food borne disease prevalence in food service establishments can be attributed to food handlers improper food preparation practices (Aklilu et al. 2015). Studies indicate that education of food handlers to improve their hygiene-related knowledge and practice is of paramount importance in the prevention and control of food borne diseases (Harris 2015).

Cross-contamination happen when food handlers who work with raw food fail to wash their hands before handling cooked food and proper cleaning of equipment and utensils such as knives or chopping boards used during the preparation of raw and cooked food (Vasconcelos et al. 2016). Intestinal parasites transmitted directly or indirectly through faeces contaminated objects like food, water, soil and finger (Marami et al. 2018)

Intestinal parasites of humans were a very diverse group of animals, ranging from single celled protozoans to multicellular worms that inhabit the gastrointestinal tract (Jallow et al. 2017). The prevalence of intestinal parasites depends on factors like poor socioeconomic condition, lack of safe and adequate water supply, poor environmental sanitation, irrigation, overcrowding, resettlement, and low altitude, lack of proper disposal of waste, noncompliance with health standards, lack of adequate washing of vegetables, and consumption of uncooked meat, the practice of personal hygiene, knowledge of disease transmission, different environmental factors and education status of the food handlers (Abossie and Seid 2014; Mama and Alemu 2016).

According to National Food Safety Standards of Iran, all food handlers must undergo parasitological stool examination prior to receiving their health certificate. Therefore all food handlers were referred to a health center medical diagnosis laboratory to get checked for intestinal parasitic infections (SHARIF et al. 2015).

Food handlers working in food service establishments with poor personal hygiene can be infected by different pathogens where they can cause fecal contamination of foods by their hands during

food preparation and which might be transmitted to the public. Therefore, a proper screening procedure for food handlers was helpful in the prevention of probable morbidity and the protection of consumer health. Due to the surviving of microorganisms and difficult to keep them clean finger nails are the most contaminating part of hand (Abate et al. 2013; Tefera and Mebrie 2014).

1.2 Statement of the problem

Food safety problems were an important challenge to the public health sector because many cases of food-borne illnesses were unreported and unrecognized. Food borne illness was a significant contributor to the burden of disease in developing countries and causes death and suffering even in the developed countries of the world (Nematian et al. 2004).

The burden of food borne illnesses was transmitted from poor hygienic practices either by food handlers or during food production. Parasitic disease results in chronic intestinal blood loss, iron deficiency anemia, diarrhea, loss of weight, abdominal pain, nausea, physical and intellectual growth retardation, vomiting, lack of appetite and abdominal distention. In children chronic hookworm disease retards physical growth which is sometimes most apparent at puberty (Abera, Biadegelgen, and Bezabih 2010; Hailegebriel 2017).

Soil transmitted helminthes were diseases caused by helminthes with direct life cycle through the soil environment to man and domestic animals (Oyebamiji et al. 2018).

As the study in tropical and sub-tropical countries soil transmitted helminthes were parasitic nematode worms causing human infection through contact with parasite eggs or larvae that thrive in the warm and moist soil. An estimated case of 1.471 billion infection and 65 000 deaths occur globally due to A.*lumbricoides.* Hookwormwas one of the most common cause of chronic infections with an estimation of 1.3 billion cases globally and 65 000 deaths annually. *T. trichiura* estimation was 1.049 billion cases of infection and 70 000 deaths annually (Mama and Alemu 2016).

Intestinal parasitosis is one of the foods borne disease in developing countries including Ethiopia. From the population of Ethiopia one third, one fourth and one in eight infected by *A. lumbricoides*, *T. trichiura* and Hookworm respectively (Lalit, Brkti, and Dejen 2015).

Federal Democratic Republic of Ethiopia Federal Ministry of Health has prioritized intestinal parasitic infection as one of the Neglected Tropical Disease (NTDs) in the National Master Plan of NTDs, to address the public health problems of NTDs. *Ascaris lumbricoides*, *Trichuris trichiura*, *Entamoeba histolytica and Giardia lambia* were the most common causes of parasitic infection (Solomon and Abraham 2016).

According to Food, Medicine and Health Care Administration and Control Authority (FMHACA), it was prohibited for any institution that engages in food production, processing, storing, distribution and transportation to hire an employee having contact with the product and who was infected with communicable disease (Gazeta 2010).

According to food standard agency food handlers fitness to work, there was a legal requirement for every person working in a food handling area to maintain personal hygiene and to wear suitable, clean and, where necessary, protective clothing. Food handlers have the responsibilities to report to the manger immediately when they were ill and wash and dry their hands with soap and water especially after using toilet in order to prevent spread of infection through food. Nails should be kept short to make hand washing easier. No person should be engaged in food handling unless he/she had food handler training card (Guidance, Advice, and Business 2009).

It was expected that all food handlers at University cafeterias to have a periodic medical checkup for food borne pathogens, trained about food hygiene and safety and training certified for ensuring hygienic food handling and preparation practice to safeguard health and well-being of students and total population (Nigusse and Kumie 2012). However, there was no report of food handling practice and hygienic status of food handlers at the study site. Therefore, the aim of this study was to fill the gap of prevalence of intestinal parasites and associated risk factors among food handlers working in Wachemo University students' cafeteria.

1.3 Significance of the Study

To provide scientific information on hygienic status of food handlers. Hand swabs of finger nail to assess the level of parasitic contamination were also used during employment to assess the level of personal hygiene practices of individuals and stool examinations also to indicate parasitic infection and sanitation problems related to food handlers. This can inform the level of supervision and training required as well as any subsequent management action.

The law requires that in all food businesses, no person suffering from, or being a carrier of a disease likely to be transmitted through food is to be permitted to handle food or enter any food-handling area in any capacity if there is any likelihood of direct or indirect contamination. Any person so affected and employed in a food business and who is likely to come into contact with food was to report immediately the illness or symptoms, and if possible their causes, to their manager or supervisor.

The information generated from this study and the suggested recommendation can also be used as an input for policy makers, nongovernmental organization, academic institutions, health planners and cafeteria administrators to take measures that mitigate the transmission of intestinal parasites in food and drinking establishments from food handlers to students and total population served by them. Therefore, information generated from this study can be used as reference for further similar studies in Ethiopia as well as in the world.

2. LITERATURE REVIEW

2.1 Intestinal Parasites

According to study on intestinal parasites among food handlers in different study areas prevalence shows that 37.2% in North Thailand (Boonjaraspinyo et al. 2013), 46.3% in Gambia (Jallow et al. 2017), 34.9% in Bandar Abbas, Southern Iran (Heydari-Hengami et al. 2018), 14.4% in Nairobi Kenya (Nematian et al. 2004), 83.1% in Swat, Khyber Pakhtunkhwa, Pakistan (Pakhtunkhwa and Khan 2017), 29.4% in Khartoum Sudan (Babiker, Ali, and Ahmed 2009), 45.3% in Addis Ababa University (Aklilu et al. 2015), 36% in Arba Minch University (Mama and Alemu 2016), 41.1% in Bahirdar town (Abera et al. 2010), 13.8% in Gondar University (Gelaw et al. 2013), 25.2% in Haramaya University (Marami et al. 2018), 33% in Jimma University (Hundaol et al. 2017), 81% in Chencha town Southern Ethiopia (Abossie and Seid 2014), 33.68% in Wolaita Sodo town (Solomon and Abraham 2016), 44.1% in Yebu town Western Ethiopia (Tefera and Mebrie 2014) and 14.5% in Northern Ethiopia Axum town (Gezehegn et al. 2017).

The study revealed that fingernail contents examined among food handlers in different study area prevalence of ova of intestinal parasites were 10.9% in Jimma town (Zewdneh 2001), 29.1% in University of Indonesia (Manan 2011), 21% in Lahore Punjab Pakistan (Ghani et al. 2016), 57.2% in Ebonyi State Nigeria (Alo, Ugah, and Elom 2013), 2% in Sohag Egypt (El-Nadi et al. 2017) and for *A.lumbercoids* 4.95 in Jimma town, 9.52 % in Lahore Punjab Pakistan, 20% in Ebony State Nigeria and 2% in Sohag Egypt.

According to study conducted in Bagalkot city, Karnataka, India on Prevalence of intestinal parasites and its associated socio-demographic factors among the food handlers show that 39 (14.7%) were tested positive for intestinal parasitic infections. Most common parasite isolated was *A. Lumbricoides* (5.3%) followed by *E. Histolytica* (1.5%), *Giardia (1.5%)*, *T. Solium* (0.8%), *T. Trichura* (0.8%) and *H. Nana* (0.8%). Mixed infections constituted 4.1%(Anjum, Kalasker, and Bhaskar 2017).

According to study conducted in Addis Ababa University among food handlers on prevalence of intestinal parasites and associated risk factors reveals that from 172 stool specimens, 78(45.3%) were positive for different intestinal parasites. The most abundant parasite was *E. histolytica/ dispar* 68 (70.8%) followed by *Giardia lamblia* 18 (18.8%), *Taenia saginata* 5 (5.2%), *Ascaris lumbricoides 2 (2.1%), Trichuris trichiura 1 (1.1%) and hookworm* 2 (2.1%). Among 78 positive

food handler's, 18(18.8%) had mixed infections. The dominant parasites among mixed infections were *E. histolytica and G. lamblia*(Aklilu et al. 2015).

According to guidelines for the evaluation of Soil-transmitted helminthiasis and schistosomiasis at community level Prevalence is high when it is >50%, moderate 20 %-< 50% and low <20% (Sturrock 1998).

2.2 Transmission risks of intestinal protozoa and helminthes infections

The risks associated with transmission of intestinal parasites are *personal hygiene practice factors, environmental factors, and social economic status and knowledge factors*.

2.2.1 Personal hygiene factors

As the study in Dire dawa University revealed that Statistical analysis of hand washing habit after toilet shows that there is a statistically significant difference in isolation rate of *Shigella* between food handlers that washed their hands with soap and water and only with water after toilet, p=0.040. Therefore, washing hands with soap and water has 88% protective effect than washing hands only with water after toilet in acquiring *Shigella* infection with 31% AOR:0.12; with a 95% CI=0.01-0.91. Statistically significant association was also seen between isolation rate of *Shigella* and finger nail status, p=0.026. Therefore chance of getting *Shigella* infection while finger nails are trimmed is 91% lesser than while having semi trimmed or untrimmed finger nails AOR=0.09; with a CI=0.01-0.7 (Tamiru et al. 2015).

Study conducted in Addis Ababa University among food handlers said that Hand washing practice before food preparation was 76%, AOR:0.15 with 95% CI = 0.06-0.38 (Aklilu et al. 2015).

According to study conducted in Arbaminch University, the practice of hand washing after toilet (p = 0.029) and before food handling (p = 0.034) was significantly associated with parasitic infection among the study participants. Food handlers who were using water only when they washed their hands after toilet had a more likely risk of infection (with 71 %) for intestinal parasites AOR:1.71, 95 % CI= 1.057–2.765 than food handlers who use water and soap and water only with 71%, AOR:1.71 with 95% CI=1.057-2.765 (Mama and Alemu 2016).

According to study conducted in Jimma, hand washing before serving food and after toilet use was 48 (51.1%) of the food-handlers were not washing their hands and among them 24 (50.0%) were tested positive for some kinds of parasites. Of the 94 food-handlers, only 40 (42.6%) trimmed

finger-nails, of whom 2 (5.0%) were positive for one or more species parasites. Of the 54 (57.4%) with untrimmed finger-nails, 24 (44.4%) were positive. Untrimmed finger-nails showed a higher prevalence than the trimmed ones and the difference was statistically ($P < 0.05$) significant (Girma et al. 2017).

According to study done in Mekelle University students' cafeteria on food hygiene practices and prevalence of intestinal parasites among food handlers the total respondents, 195 (70.4%) of them stated that they have a habit of hand washing with soap and water with 85%, AOR: 0.15 with 95% CI = 0.06-0.38. One hundred two (36.8%) respondents had medical checkups and 117 (42.2%) respondents had a history of de-worming either once (73.5%), twice (17.9%) or thrice (4.3%), in the past one year prior to this study. Only 34(12.3%) respondents were certified with six months formal food handler's training program from different training centers. Among these, 28 (82.4%) of them reported that the training was useful for the provision of food safety in their work places 202 (72.9%) used soap when they washed their hands before preparing food in any situations. The majority, 245(88.4%) reported that they kept their finger nail cut short (Nigusse and Kumie 2012).

According to study conducted in Aksum town in Northern Ethiopia 310 (77.5%) reported that they always wash their hands before food preparation, 295 (73.8%) always use soap and water after visiting toilet and 303 (75.8%) wash their hands after touching dirty material and different body parts (Gezehegn et al. 2017).

According to study conducted on prevalence of intestinal parasites and associated risk factors among asymptomatic food handlers in Haramaya University showed that lack of hand washing after use of toilet with soap and water with 65%, AOR=2.43 with 95% CI=1.22-4.86 and untrimmed fingernails [AOR: 3.31, 95% CI: 1.99, 5.49] were significantly associated with IPIs (Marami et al. 2018).

Cross-Sectional Study on the prevalence of intestinal parasites and associated risk factors in Teda Health Centre, northwest Ethiopia indicate that Swimming and less shoes wearing habits showed a statistically significant association with prevalent *S. mansoni* and Hookworm infections, respectively. The prevalence of *S.mansoni* was higher in study participants who had swimming habit (18.9%) than who did not. This might indicate the presence of infested water bodies in the study area (Abate et al. 2013).

2.2.2 Socio demographic and economic factors

A cross sectional study conducted in Prevalence of intestinal parasitic infections and their relation with socio-economic factors and hygienic habits shows that intestinal parasitic infection prevalence was not significantly associated with age and gender (p=0.053), with years of service (p=0.086) (Nematian et al. 2004).

Study conducted in India revealed that among food handlers, majority of the food handlers were males 73.7%. Most of food handlers (44.7%) are young between 21-35 years age group, 59.4% were from urban area, 75.6% were literates and waiters constituted largest group with 35.3%. About 46.3% were having work experience between 1-5 years and majority (66.2%) of subjects were married (Anjum et al. 2017).

Study conducted in Jimma University Specialized Hospital cafeteria Southwest Ethiopia on intestinal parasites show that among 94 food-handlers participated in the study area 42.5% waiters, 39.4% cookers and 18.1% cleaner of utensils. The majority 74.5% (70/94) of food-handlers had 5 or less years of work experience (Girma et al. 2017).

According to study in Iran the prevalence of intestinal parasitic infection in Iran was 10.4%. In this study statistically significant difference in the prevalence of parasite infection was reported between males and females (p=0.024), while there was no difference among different educational groups (Heydari-Hengami et al. 2018).

2.2.3 Environmental factors

According to study done in Tanizania on determination of hygienic practice and status of food Handlers in catering premises, equipment and utensils like knives or chopping boards used with uncooked products such as raw meat or poultry can become contaminated with pathogens during food preparation. Cross-contamination can also happen if food handlers who work with raw food fail to wash their hands before handling cooked food (Punsawad et al. 2018).

As a result of poor sanitation problems, water sources were contaminated by human's faeces. So that parasites are easily transmitted by faeco orally. The parasite eggs and cysts were adhering to dust, utensils, finger nails, door handles and currency notes and coins. Flies and cockroaches may serve as vectors by ingesting the cysts and/or eggs present in faeces and depositing them in food or mechanically carry them on their (Nematian et al. 2004).

According to this study factors affect transmission of helminthiasis were improper cleaning after using the latrine, bad habit at defecation, use of unclean soil contaminated feeding equipment, eating unwashed vegetables and fruits and other utensils (Punsawad et al. 2018). Most intestinal parasites were transmitted through ingestion of fecal contaminated food or water. Shoes prevent larvae from penetrating feet (Babiker et al. 2009).

According to study from Mekele University student's cafeteria on food hygiene practice and intestinal parasites an adequate potable water supply, with appropriate facilities for its storage and distribution ensure the safety of food in establishments. Adequate drainage and waste disposal system, utensils and equipment cleaning facilities with hot and cold water supply are crucial for food safety practice (Nigusse and Kumie 2012).

2.2.4 Participants knowledge of intestinal parasites

Study conducted in high end hotels in Nairobi Kenya on Prevalence of intestinal parasitic infections among food handlers 92.3% were knowledgeable about the intestinal parasite and 7.7% were not aware of the intestinal parasites. Among those asked about infectivity of intestinal parasites 75.8% said infected. Of 75.5% of study participants strongly agreed that washing hands before eating food was very important. 70.1% were not aware on the purpose for the routine medical examination (Nematian et al. 2004).

Study conducted in south western Nigeria on knowledge, practice and attitude reveal that among the study participants 62.6% were have knowledge of parasitic worms while 88.4% do not actually know how to avoid getting infected with worms. Analysis showed that a significantly lower proportion of respondents (11.6%) reported that clean water and clean environment could prevent transmission of STH (Oyebamiji et al. 2018).

According to the study from Dangila town North West Ethiopia on factors affecting food handling Practices among food handlers. Food handlers who had good knowledge were 1.69 times more likely to have good food handling practices compared to those who had poor knowledge AOR: 1.69, 95% CI = 1.05-2.73.This finding was in line with the findings in Mekelle with AOR: 3.61, 95% CI = 1.51-8.65. From the participants 289 (71.2%) of the respondents had poor knowledge score on food handling Practices. Most of food handlers 361(88.9%) had heard about food borne diseases of which 117 (32.4%) had a good knowledge. Among food handlers who believed that

personal hygiene prevents food borne disease of which 117 (28.6%) have a good knowledge (Tessema, Gelaye, and Chercos 2014).

2.3 Conceptual Frame Work

The socioeconomic, personal hygiene practice, knowledge and environmental risk factors that might lead to the occurrence of intestinal parasites among food handlers were interlinked to cause illness and that one factor alone might not lead to an intestinal parasites but a combination of factors can cause disease.

Conceptual frame work shows how dependent and independent variables were related each other. The dependent variables are prevalence of intestinal parasites. The dependent variables were affected by independent variables like socio demographic/economic conditions of the food handlers like age, marital status, sex, food safety and hygiene training, income, educational status and occupation, personal hygiene practice like hand washing practice, , wearing protective clothes, hand washing habit, fingernail status, sharing of knife ,swimming ,eating raw/ cooked ,Sickness reporting and medical checkup ,environmental risk factors like water source, availability of water and soap ,toilet facility, shower facility, hand washing facility and knowledge of the food handlers were used to predict intestinal parasites.

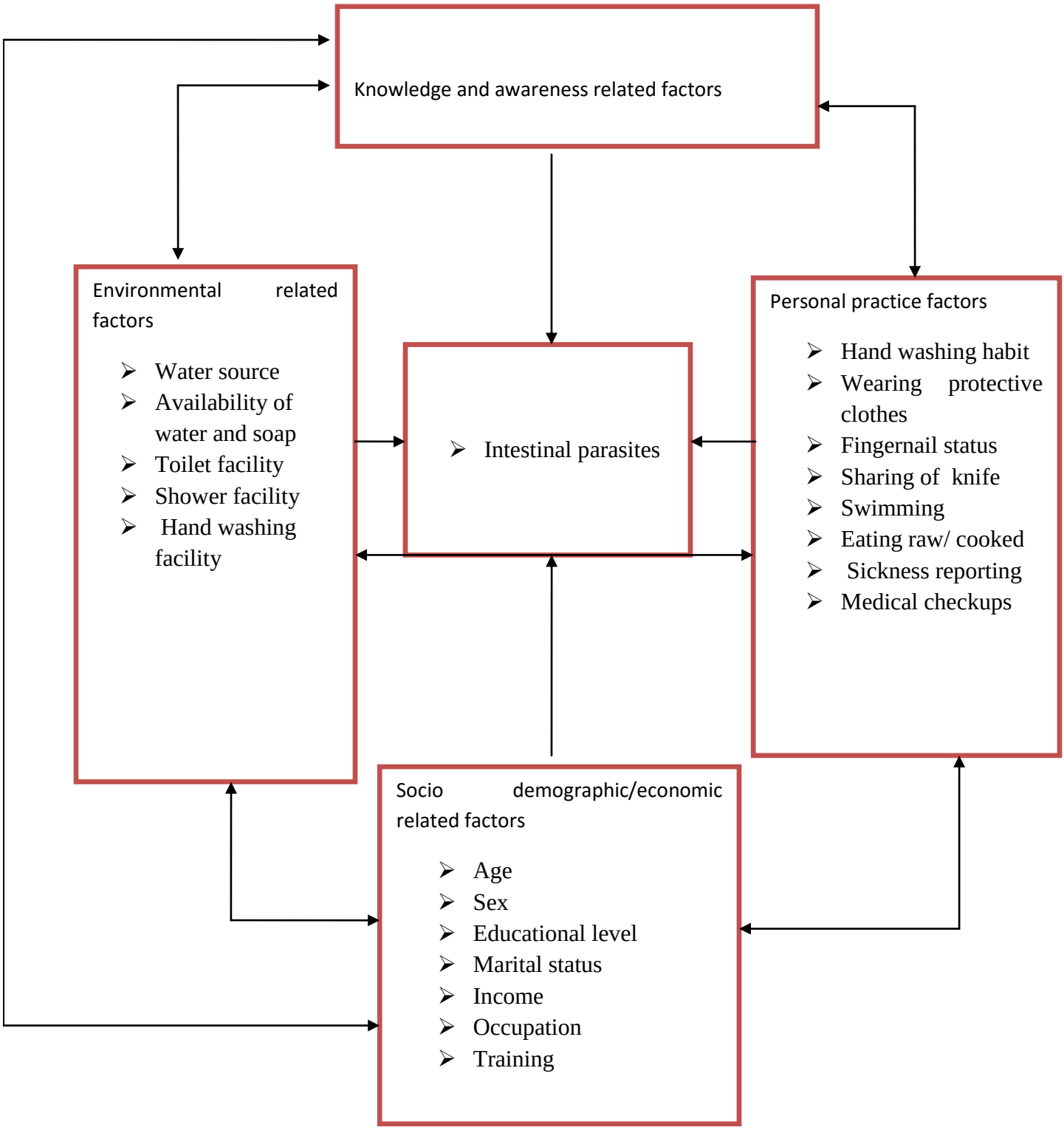

Figure 1: Conceptual Frame work that shows the relationship of dependent and independent variables among food handlers of Wachemo University, students' cafeteria.

3. OBJECTIVE

3.1 General objective

To determine prevalence of intestinal parasitic infections and predisposing factors among food handlers working in Wachemo University students' cafeteria, Southern, Ethiopia.

3.2 Specific objectives

To determine prevalence of intestinal parasites.

To determine ova of parasite from finger nail swabs.

To assess risk factors for the intestinal parasitic infections among food handlers.

4. MATERIALS AND METHODS

4.1 Study area

Wachemo University was one of public higher educational institutions which is found in Southern Nations, Nationalities, and Peoples' Region (SNNPR) and established in 2009. It was located 230km south west of Addis Ababa. Occupying total area of 200 hectares and it was 3km away from the center of Hosanna town. The topographic feature of the town is of both plain and plateau lands, and this has contributed to its natural scenery. It was committed to Excellence in teaching, research, and service for the benefits of the citizen of Ethiopia and beyond. The university started its functions in 2012 admitting 538 students and currently the university had admitted over 18000 students in regular and continuing education programs. The University comprises two campuses namely Main and Durame campus. Based on the information collected from the human resource management during the study period a total 520 individuals were serving as food handlers throughout Wachemo University cafeterias. At present, the University runs both undergraduate and post graduate programs in two campuses (Demmelash 2018).

4.2 Study period

The study was conducted from Feb-March /2019.

4.3 Study design

A Cross-sectional study design was conducted.

4.4 Population

4.4 .1 Source population

All food handlers who had direct contact in food preparation and handling regardless of their employment status either permanent or contract employee during the study period in students' cafeteria of Wachemo University.

4.4.2 Study population

All randomly selected food handlers who had direct contact in food preparation and handling regardless of their employment status either permanent or contract employee within students' cafeteria of Wachemo University.

4.4.3 Study Subjects

Study subjects were food handlers who were selected by simple random sampling technique from study population who met inclusion criteria within in Wachemo University students' cafeteria.

4.5 Eligibility criteria

4.5.1 Inclusion criteria

All food handlers who had a direct contact with foods and drinks were included in the study.

4.5.2 Exclusion criteria

Food handlers who had diarrhea, fever, taking antibiotics, antihelminthics were excluded from the study. Food handlers who had taken annual leave during data collection time or those who had received medical treatment for any intestinal disease within the past 2 to 3 weeks prior during data collection were excluded from the study.

4.6 Sample size determination and sampling technique

4.6.1 Sample size determination

For objective one: sample size was determined using sample size determination for estimation of single population proportion formula (Mama and Alemu 2016).

$$n=\left[Z\alpha/2\right]^2 * p\left[1-p\right]/d^2$$

Where

Z = 95% confident level.

Z α/2 = Standard normal deviation (1.96) at a 95% confidence interval.

P = Estimated proportion of parasite prevalence rate (p=36%) from previous study from Araba minch University (Mama and Alemu 2016).

d= Margin of sampling error tolerated between the sample and population 5%.

n= (1.96)2 0.36(0.64) = 354

(0.05)2

For objective two: sample size was determined using sample size determination for estimation of single population proportion formula.

$$n=[Z\alpha/2]^2 * p[1-p]/d^2$$

Where

Z = 95% confident level.

Z α/2 = Standard normal deviation (1.96) at a 95% confidence interval.

P = estimated proportion of parasite prevalence rate (p=10.9%) from previous study from Jimma University (Zewdneh 2001).

d= Margin of sampling error tolerated between the sample and population 5%.

n= (1.96)2 0.109(0.891) = 149

(0.05)2

For objective three: sample size was determined using EPI INFO version 7 by taking variables that have significant association with intestinal parasites in studies and considering the assumptions as indicated in the following Table.

Table 1 Sample Size for Factors Associated with Intestinal Parasites among Food Handlers in Wachemo University Student's Cafeteria, Southern Ethiopia, (2019).

s.no	Variable	Assumptions					Sample size	reference
		Confidence level (%)	Power (%)	Ratio(Unexposed : exposed)	% outcome in an exposed group	OR		
1	Finger nail status	95	80	1	54.27	3.68	106	(Abera et al. 2010)
2	Hand washing	95	80	1	34.88	10.6	36	(Hailegebriel 2017)

Maximum sample size from the calculated for the above three objectives was 354. Since the source population was less than 10000, finite population correction formula was used.

$nf = n/ (1+n/N)$

Where nf= final sample size

n= calculated sample size

N= total number of food handlers at Wachemo University cafeteria.

$nf =354/(1+354/520) = 212$

Finally by adding 5 %(11subjects) for non-respondent **final working sample** was 223.

4.6.2 Sampling technique

To select representative groups from 520 number of total food handlers a proportional allocation were used to assign sample value for each cafeteria and simple random sampling technique were used and food handlers were selected randomly by lottery method from the roster lists of food handlers which was obtained from cafeteria office of Wachemo University.

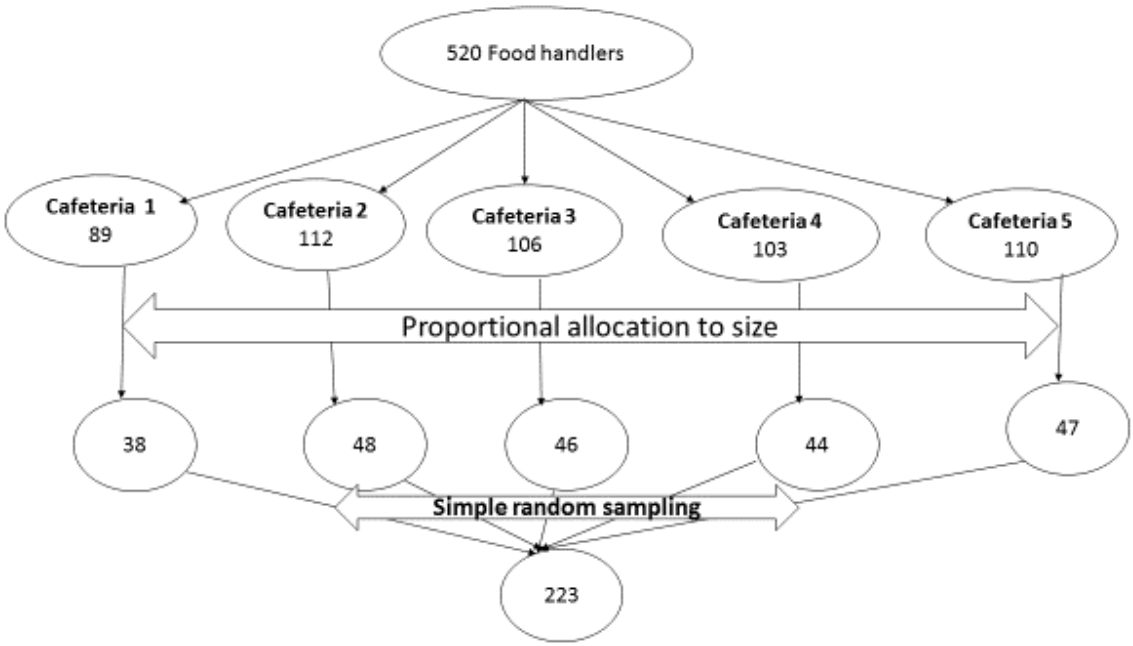

Figure 2 Flow chart for sampling technique for food handlers in Wachemo University, Southern Ethiopia, (2019) (n=223).

4.7 Methods of data collection

4.7.1 Questionnaire

The benefits of study were explained for food handlers before interviewing, collection of stool and ova specimen by laboratory technologist and data collectors (Annex I and Annex II).Semi-structured questionnaires were used to collect data. A total of 212 copies of predesigned questionnaires were used to collect data related to socio-demographic/economic characteristics, knowledge on parasite transmission, environmental and personal hygiene practices of food handlers and related risk factors were collected by face to face interview using pre tested questionnaire and observational check lists by trained Nurses and Health Information

Administrator professionals/HIA/(Annex III). The questionnaire were adopt from the World Health Organization (WHO) food safety checklist and literatures.(Abera and Nibret 2014; Gezehegn et al. 2017; Mama and Alemu 2016) Data on fingernail trimming, wearing hair cap, gown and whether the participants were on barefoot or not were recorded by a simple observation.

4.7.2 Collection and examination of stool and ova specimen

4.7.2.1 Collection of stool and ova specimen

Selected food handlers were provided with a labeled clean stool cup, applicator stick and a plain paper. The stool cup had a code number; the code number of the container was recorded in a laboratory report format (Annex IV). This was so as to avoid the accidental exchange of specimens among food handlers. The food handlers were instructed that once they go to the latrine, defecate on a piece of paper provided, to avoid contamination from the toilet environment and then using an applicator stick they pick up a portion of the stool and put it into the clean plastic container provided and deliver it. According to list of names with their corresponding code numbers, stool specimen will be collected. The number on the container was compared with the number recorded when they provided the container to check if it was the right container for her/him.

The specimen from hands swabs was collected from the hands using moisten sterile cotton swab, with (0.9% w/v) physiological saline. The swab was kept quickly into its container and sealed with unique code number.

The collected samples were transported immediately for examination to NigestElieniMahammed Memorial Referral Hospital of Wachemo University and Gimbichu Primary Hospital.

4.7.2.2Examination of stool and ova specimen

About 2mg of stool was picked with applicator stick and emulsified in a drop of normal saline (0.85% NaCl) at the one end of a clean, non-scratched glass slide, and the same size of stool in Lugol‟s iodine at the opposite end of the same glass slide. Then cover slip was placed on both preparations and scanned under 10× and 40× objective lenses of a light microscope for detection of intestinal protozoan trophozoite, cysts and ova of nematodes. Briefly, formol ether concentration technique was carried out as follows: about 1gm (pea size) of fresh stool specimens were emulsified in 8ml of formol water. The resulting suspension was filtered through three layers of wet cotton gauze in a funnel into a centrifuge tube and about 4ml of diethyl acetate is added. The centrifuge tube was shaken vigorously and centrifuged at 750-1000g for 1 minute. Then after

the supernatant poured off the sediment and slightly shaken and one drop was added at the center of clean slide, non-scratched with free glass slide. Cover slip was applied on the preparation and examined in the same way as explained earlier. Small drop of Lugol‟s iodine was allowed to run under the cover slip to observe the characteristic features of cyst. Dysenteric (watery) stools were examined before solid stools. This was necessary because protozoan trophozoites were more likely to be present in watery stool; and trophozoites had a shorter survival time in feces than cysts. The different intestinal parasites identified were recorded on laboratory investigation result recording form (Annex IV).

4.8 Study Variables

4.8.1 Outcome Variable:

The outcome variable was the occurrence of intestinal parasites among food handlers.

4.8.2 Predictor Variables

Socio demographic/economic factors

- Age
- Sex
- Educational level
- Marital status
- Income
- Occupation
- Training
- Working condition

Environmental factors

- Water source
- Availability of water and soap
- Toilet facility
- Shower facility
- Farming activities
- Separate hand washing facility

Personal practice factors

- Hand washing habit
- Wearing protective clothes
- Fingernail status
- Sharing knife to cut
- Eating raw or cooked
- Sickness reporting
- Medical checkups

Knowledge and awareness factor

- Information
- Sources
- Transmission
- Prevention
- Treatment
- Contaminations

4.9 Operational Definitions

Food handler: was a person performing under contractual agreement or permanent employee who handles, prepares, serves or who comes in contact with eating or cooking utensils or other equipment used in the handling, preparation and service.

Shoes: any kind of shoes that used by food handlers not to walk on bare foot.

Clean apron: is protective cloth which is washed and used by food handlers to serve students.

Knowledge: To assess the level of knowledge, respondents were asked 13 questions questionnaire and those who scored ≤ the mean value were considered as having poor knowledge and those who scored > the mean value were considered as having good knowledge.

Always: washing hands with soap and water frequently after toilet use, body and raw material touching and before food preparations three times a day.

Sometimes: not washing hands with soap and water frequently after toilet use, body and raw material touching and before food preparations three times a day.

Finger nail status: Assessing food handlers both hands of finger nail whether trimmed or untrimmed by observing.

Hygiene: the promotion of cleanliness to prevent spread of diseases.

4.10 Data analysis

Data was edited, cleaned, entered and analyzed using EPI data 3.3.1 and statistical package for social science (SPSS) version 20. Descriptive analysis like frequencies and mean were used. Odds ratio was analyzed to measure the strength of the association. Bivariate analysis were used, crude and adjusted odds ratio with 95 % CI was calculated for statistical significance tests. The goodness of fit model was checked by Hosmer Leme show statistic and p-value greater than 0.05 was considered as a fit model. Variables with significant at P<0.25 (Marami et al. 2018) in a bivariate analysis were considered for multivariate analysis through multiple logistic regression model to look their relative effect on the outcome variable by controlling other possible confounding factors and Significantly associated factors were selected using backward stepwise (Likelihood ratio) method. P-value with <0.05 was considered for indicating statistical association. Lastly the result was presented by using tables, graphs and described by narration.

4.11 Data quality control

To ensure the quality of data standardization of the study tools and procedures was done. The English version of questionnaire was translated to Amharic and Hadiyegna language and back translated to English by translators who are blind to the original questionnaire. Training for all research team members consists of 2 days of classroom instruction and practice and 1 day of pretesting in Hossana Health Science College among food handlers of cafeteria in all research procedure including interviews and stool and ova specimen collection in 5% of the sample size one week prior to actual data collection period. Some of the variables were corrected after pretesting tools like medications which is similar with medical checkup, job responsibilities, frequency of hand washing. Stool and ova sample collection and investigation was conducted according to standard procedures. Microscopic reading was made by the first author and a result was confirmed by him.

During data collection at each selected food handlers research team supervisor were supervised all steps of data collection including the interview and stool and ova specimen collection. Upon completion of data collection at each food handlers the PI was review the entire data collection form to ensure completeness and accuracy.

4.12 Ethical considerations

Ethical clearance was obtained from Jimma University Ethical Committee. Official letter of co-operation from the above organization was written to Wachemo University. Prior to the date of data collection, translated consent forms was forwarded to the food handlers requesting for the food handlers participation in the research as well as highlighting the need for collecting stool and ova samples from the food handlers. Food handlers were requested to return completed consent forms. A written informed consent was signed by participants. When food handlers were illiterate, they were consented by their thumbprint after verbal consent by the interviewer during data collection. The procedure was not harmful to the subjects. Those who tested positive for the parasites were recommended for treatment. Additionally confidentiality of all the information was assured.

4.13 Dissemination and utilization of result

The findings will be disseminated or presented to Jimma University Institute of Health, Faculty of Public Health Department of Environmental Health Sciences and Technology. After approval it will be disseminated especially for Wachemo University and also it will be published in scientific journal. A feedback session for participating food handlers and key stakeholders will held at the end of the research to present and discuss the findings and to jointly draft a plan of action.

5 RESULTS

5.1 Socio-demographic and economic data

From a total of 212 food-handlers working at Wachemo University and recruited to participate in the current study, 135 (63.7%) were females and 77 (36.3%) were males with response rate of 95%. The mean age of the respondents was 31.51 years. Regarding their educational status most of the study participant's 102 (48.1%) were attended grades nine to ten (Table 2).

Among 212 food-handlers participated in this study, 89(42%) were waiters. According to their marital status 109(51.4%) were single and 101(47.6%) were married. Related to monthly income, most of the study participants (71.2 %,) earn less than 1500 birr with very few (5.2%) earning more than1801birr.

Regarding to training, 37(17.5%) were trained and only 12(5.7%) were certified, 175(82.5%) were not trained and 200(94.3%) were not certified (Table 2).

Table 2 Socio Demographic and Economic Characteristic of Food Handlers in Wachemo University Students' Cafeteria, Southern Ethiopia, (2019) (n=223).

Variable	Category	Frequency [n (%)]
Sex	Male	77 (36.3)
	Female	135 (63.7)
	<20	13 (6.1)
Age	21-30	94 (44.3)
	31-40	87 (41)
	>40	18 (8.5)
Educational status	1_4	13 (6.1)
	5_8	57 (26.9)
	9_10	102 (48.1)
	11_12	28 (13.2)
	>12	12 (5.7)
Job responsibility	Waiter	89 (42)
	Cooker	80 (37.7)
	Equipment collector	19 (9)
	Dish washer	24 (11.3)
Marital status	Single	109 (51.4)
	Married	101 (47.6)
	Divorced	1 (0.5)
	Widowed	1 (0.5)
Income per month	<1500 ETB	151 (71.2)
	1501-1800 ETB	50 (23.6)
	>1801 ETB	11 (5.2)
Training	Yes	37 (17.5)
	No	175 (82.5)
Training certificate	Yes	12 (5.7)
	No	200 (94.3)
Working condition	Permanent	17 (8)
	Contract	195 (92)

5.2 Personal hygiene related factors of food handlers

Of the total 212 respondents, 99 (46.7%) reported that they trimmed their finger and 113(53.3%) not. For cutting raw and cooked foods 204(96.2%) were not using common knife, 209(98.6%) were wear clean apron, 210(99.1%) had a habit of wearing shoes and 75(35.4%) had a habit of reporting to the mangers when sick. After using toilet facility 97(45.8%) and 115(54.2%) wash their hands always and sometimes respectively. Similarly, 163(76.9%) wash their hands before touching foods, 66(31.1%) wash their hands with water and soap while 157(74%) wash their hands only with water, while 146(68.9%) were not washing their hands with soap and water, Moreover 209 (98.6%) were washing fruits and vegetables before eating (Table 3).

Table 3 Personal Hygiene Practices of Food Handlers in Wachemo University Students' Cafeteria, Southern Ethiopia, (2019) (n=223).

Variable	Category	Frequency [n (%)]
Finger nail status	Trimmed	99 (46.7)
	Untrimmed	113 (53.3)
Sharing knife	Yes	8 (3.8)
	No	204 (96.2)
Wearing clean apron	Yes	209 (98.6)
	No	3 (1.4)
Wearing hair cap	Yes	211(99.5)
	No	1(0.5)
Shoe wearing habit	Yes	210 (99.1)
	No	2 (0.9)
Reporting when sick	Yes	75 (35.4)
	No	137 (64.6)
Hand washing after using toilet	Always	97 (45.8)
	Sometimes	115(54.2)
Wash hands before touching food	Yes	163(76.9)
	No	49 (23.1)
Washing hands with soap and water	Yes	66 (31.1)
	No	146 (68.9)
Washing hands with water only	Yes	157 (74)
	No	51 (26)
Washing fruits and vegetables before eating	Yes	209 (98.6)
	No	3 (1.4)
Follow medical check up	Yes	75 (35.4)
	No	137 (64.6)
Washing cloth in the river	Yes	210 (99.1)
	No	2 (0.9)

5.3 Environmental related factors of food handlers

From the total respondents, 21(9.9%), 140(66%), 51(24.1%) were obtained vegetables and fruits from farm, market and farm and markets, respectively. According to this study 21(9.9) were participated in farming activities like digging and weeding but 191(90.1%) did not participated. All 212 (100%) respondents were users of private tap water, and 35(16.5) had shower facility.

All of the respondents had a toilet facility in their establishment of which, 187 (88.2%) had pit latrine and 25(11.8%) had water flush type of toilet. Among those toilets, 87(41%) had separate hand washing facility while 125(59%) hadn’t separate hand washing facility (Table 4).

Table 4 Environmental Related Risk Factors of Food Handlers in Wachemo University Students' Cafeterias, Southern Ethiopia, (2019) (n=223).

Variable	Category	Frequency [n (%)]
Source of water		
	Tap water	212 (100)
Farming activity		
	Yes	21 (9.9)
	No	191 (90.1)
Toilet facility		
	Yes	210 (99.1)
	No	2 (0.9)
Toilet facility types		
	Pit latrine	187 (88.2)
	Water flush type	25 (11.8)
Hand washing facility		
	Yes	87 (41)
	No	125 (59)
Shower facility		
	Yes	35 (16.5)
	No	177 (83.5)
Vegetables and Fruits		
	Farm	21 (9.9)
	Market	140 (66)
	Farm and Market	51 (24.1)

5.4 **Knowledge related factors of food handlers**

Majority of food handlers, 142 (67%) have good knowledge score on food handling and remaining 70 (33%) food handlers had poor. From the study participants those of who had got good knowledge 29% were infected with parasitic infection where as who got poor knowledge 31% were also infected with parasitic infection.

Table 5 knowledge level of food handlers in Wachemo University Students' cafeteria, southern Ethiopia (2019) (n=223).

Knowledge category	Positive		Negative	
	Number	%	Number	%
Good	41	29	101	71
Poor	22	31	48	69

5.5 Prevalence of intestinal parasites

From the total 424 samples (212 stool and 212 finger-nail contents) examined, 63 (29.7%) with 95% CI (24.1-35.2) were positive for one or more parasites from stool and 12(5.6%) with 95% CI (0.9-12.3) were positive for one or more parasites from finger nail swab examinations.

A total of five genera of intestinal parasites were identified including, *A. lumbricoides*, Hookworm, *G. lamblia*, *T. saginata* and mixed infection like *A. lumbricoides*, Hookworm, *G. lamblia.*

The most prevalent parasite identified in stool specimens were *A. lumbricoides* (12.7%) followed by Hookworm (6.6%), *G. lamblia* (4.7), *T. saginata* (2.4%) and mixed infection like *A. lumbricoides*, Hookworm, *G. lamblia (3.3%).*

From the finger nail examined positive cases, 6 of them harbored ova of *A. Lumbricoides*, 3 had ova of *G. lamblia*, 1 had ova of *T. saginta* and 2 had harbored mixed ova of hookworm and *G. lamblia (*figure 4).

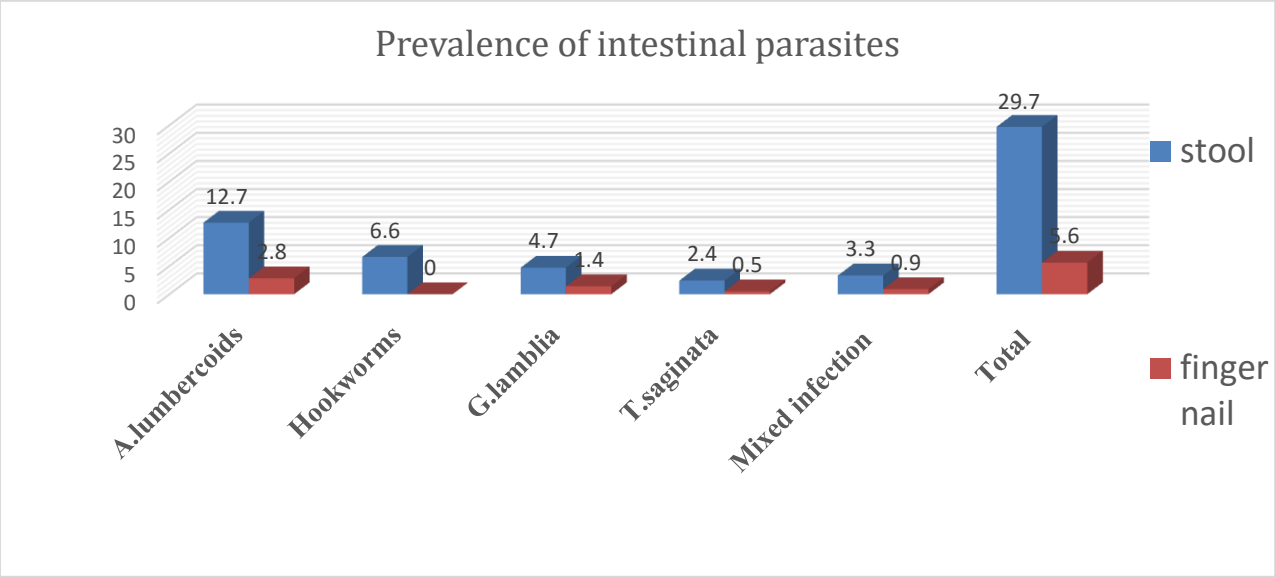

Figure 3 Type and Prevalence of Intestinal Parasites Isolated From Stool And Finger Nail Swabs of Food Handlers in Wachemo University Students' Cafeteria ,Southern, Ethiopia (2019) (n=223)

5.6 Intestinal parasitic infections and associated risk factors

As shown in Table 5, different factors were assessed for possible association with intestinal parasitic infection among the study participants. From the binary logistic regression analysis 9 variables met the criteria (p-value < 0.25) to select variables for multivariate analysis.

The logistic regression analysis result showed that the odd of being positive for intestinal parasites among male food handlers had a less likely to occur with protective effect 93% [AOR: 0.070; 95% CI (0.019-0.258)] compared to female food handlers.

Food handlers who attended primary education from 5-8 had a less likely risk of being infected with intestinal parasites with protective effect 94.5% [AOR: 0.055, 95% CI (0.007-0.413)] as compared to those food handlers who were secondary school and above.

The extent of intestinal parasitic infection was less likely to occur with a protective effect of 67%, among food handlers who had trimmed their finger nail [AOR: 0.330; 95% CI (0.113-0.965)] compared to those who had not trimmed their finger nail.

The odd of being positive for intestinal parasitic infection was less likely to occur with a protective effect 66.8%, among food handlers who washed their hands after toilet use with soap and water [AOR: 0.332; 95% CI (0.125-0.884)] than those who did wash with water only after toilet use(Table 6).

Among those nine variables four variables (sex, education, finger nail and hand washing by using soap and water) were significantly and negatively associated with parasitic infection (P-value < 0.05).

Table 6 Bivariate Analysis of Risk Factors Associated with Prevalence of Intestinal Parasites among Food Handlers in Wachemo University Students' Cafeteria, Southern Ethiopia, (2019) (n=223).

Variable	Positive		Negative		COR	95%CI	P-Value
	num	%	num	%			
Sex							
Male	10	13	67	87	0.251	0.109-0.488	0.000
Female	53	39	82	61	1		
Educational Status							
Grade 1-4	5	38	8	62	0.320	0.049-2.108	0.236
Grade 5-8	39	68	18	32	0.092	0.018-0.465	0.004
Grade 9-10	13	13	89	87	1.369	0.269-6.960	0.705
Grade 11-12	4	14	24	86	1.200	0.189-7.638	0.847
Grade >12	2	17	10	83	1		
Income Level							
<1500 ETB	39	26	112	74	2.393	0.692-8.282	0.168
1501-1800 ETB	19	38	31	62	1.360	0.364-5.075	0.648
>1801 ETB	5	45	6	55	1		
Job Responsibilities							
Waiter	20	22	69	78	1.725	0.645-4.614	0.277
Cooker	22	28	58	72	1.318	0.495-3.514	0.581
Equipment Collector	13	16	6	84	0.231	0.064-0.836	0.026
Dish washer	13	45	16	55	1		
Training status							
Yes	15	41	22	69	1.865	0.865-3.764	0.116
No	48	27	127	73	1		
Finger nail Status							
Trimmed	18	18	81	82	0.336	0.178-0.633	0.001
Untrimmed	45	39	68	61	1		
Medical check ups							
Yes	38	33	76	67	1.46o	0.803-2.656	0.215
No	25	25	73	75	1		
Frequency of hand washing							
Always	23	24	74	76	0.583	0.318-1.067	0.080
Sometimes	40	35	75	65	1		
Washing with soap and water							
Yes	15	23	51	77	0.600	0.307-1.175	0.136
No	48	33	98	67	1		

Table 7 Multivariate Analysis of Risk Factors Associated with Prevalence of Intestinal Parasites among Food Handlers in Wachemo University Students' Cafeteria, Southern Ethiopia, (2019) (n=223)

Variable	Positive		Negative		AOR	95%CI	P-Value
	num	%	num	%			
Sex							
Male	10	13	67	87	0.070	0.019-0.258	0.000*
Female	53	39	82	61	1		
Educational Status							
Grade 1-4	5	38	8	62	1.086	0.034-4.391	0.940
Grade 5-8	39	68	18	32	0.055	0.007-0.413	0.005*
Grade 9-10	13	13	89	87	1.978	0.305-12.835	0.475
Grade 11-12	4	14	24	86	3.297	0.377-28.830	0.281
Grade >12	2	17	10	83	1		
Job responsibilities							
Waiter	20	22	69	78	2.504	0.705-8.891	0.156
Cooker	22	28	58	72	2.201	0.664-7.292	0.197
Equipment collector	13	16	6	84	0.236	0.047-1.187	0.080
Dish washer	13	45	16	55	1		
Finger nail status							
Trimmed	18	18	81	82	0.330	0.113-0.965	0.043*
Untrimmed	45	39	68	61	1		
Washing with soap and water							
Yes	15	23	51	77	0.332	0.125-0.884	0.027*
No	48	33	98	67	1		

***significant at p-value < 0.05**

6 DISCUSSION

Food handlers may be carrying a wide range of intestinal parasites and have been implicated in the transmission of many infections to the public in the community and to students in University. The spread of disease via food handlers was a common and persistent problem worldwide. Therefore, this study was undertaken to determine prevalence of intestinal parasites by taking both stool and fingernail content samples among food handlers of Wachemo University Students' Cafeteria, Southern Ethiopia.

Examination of stool specimens showed a greater prevalence of intestinal parasites than their counter fingernail contents. In this study, the overall prevalence of intestinal parasites among food-handlers was 29.7% which was similar with the study conducted in Wolaita Sodo town of Ethiopia 33.68% (Solomon and Abraham 2016), Haramaya University 25.5% (Marami et al. 2018), Jimma University 33% (Girma et al. 2017) and Khartoum Sudan 29.4% (Babiker et al. 2009).

The present prevalence was higher than 13.8% in Gonder University (Gelaw*et al.*, 2013), 14.5% in Northern Ethiopia Axum town (Gezehegn et al. 2017), 14.4% in Nairobi Keniya (Palmieri et al. 2014) and 14.7% in Bagalkot city Karnatak, India (Anjum et al. 2017).

It was lower than 45.5% which was from Addis Ababa University (Aklilu et al. 2015), 36% from Arabaminch University (Mama and Alemu 2016), 41.1% in Bahirdar University (Abera et al. 2010), 37.2% in North Thailand (Cl and Fca n.d.), 46.3% in Gambia (Jallow et al. 2017), 83.1% in Swat, Khybern Pakhtun Khwa, Pakistan (SHARIF et al. 2015), 44.1% in Yebu town Westren Ethiopia (Tefera and Mebrie 2014), and 81% in Chencha town, Southern Ethiopia (Abossie and Seid 2014).

The differences might be due to differences in climate, geographical location and socio-demographic features including poverty and overall hygienic status of the populations studied, lacking proper housing, safe water supplies and hygienic waste disposal systems (Khan et al. 2017; Lalit et al. 2015; Nematian et al. 2004).

Therefore, regular evaluation of food handlers for their health status, including screening for the prevalence of parasites is important and helpful in the prevention of probable morbidity and the protection of consumer health.

The predominant parasite identified in the present study was *A. lumbricoides* with a prevalence of 12.7% followed by Hookworm *(*6.6%). This was not similar with the finding of a study conducted in Haramaya University in which *A. lumbricoides* (14.3%) and Hookworm (7.6%) were predominant parasite reported (Marami et al. 2018). The occurrence of parasitic infections is indicator of fecal pollution of soil and domestic water supply around homes due to poor sanitation and improper sewage disposal (Besufikad Belachew 2017).

The study revealed that out of the total of fingernail contents examined 5.6% were found to be positive for ova of intestinal parasites, which was consistent with 10.9% in Jimma town (Zewdneh 2001) and 2% in Sohag, Egypt (El-Nadi et al. 2017), lower than 29.1% in Side walk food vendors, university of Indonesia (Manan 2011), 21% in Lahore, Punjab Pakistan (Ghani et al. 2016) and 57.2% in Ebony State ,Nigeria (Alo et al. 2013).

This difference might be due to the poor personal hygiene and environmental sanitation, lack of supply of safe water, poverty and ignorance of health promotion practices in the study as compared to the practice elsewhere (Jallow et al. 2017; Mama and Alemu 2016).

The predominant parasite identified in finger nail examination of ova parasites in present study was *A. lumbricoides 2.8%* which was lower than 4.95% reported from Jimma town (Zewdneh 2001), 9.52% from Lahore, Punjab Pakistan (Ghani et al. 2016) and 20% from Ebony State, Nigeria (Alo et al. 2013) and in agreement with 2% of Sohag,Egypt (El-Nadi et al. 2017).

This could be due to high survival time of infective eggs in the contaminated environment indicating improper faecal disposal. The presence of cysts and ova of parasites in the fingernail contents indicate contamination of the fingernail as a potential sources transmission for intestinal parasites (Haile, Abera, and Dana 2017). So that food handlers' finger nails were contaminant part of the hand due to the surviving of microorganisms and difficult to keep them clean and it should be trimmed and washed regularly.

The analysis in this study indicated that female food handlers were at high risk of intestinal parasite infection as compared to male food handlers (AOR: 0.070; 95% CI (0.019-0.258). It disagrees with the study conducted in Intestinal helminthic infections among elementary students of Babile town, eastern Ethiopia in which male students (AOR: 0.61;95% CI(0.39-0.96) were infected than female students (Tefera, Mohammed, and Mitiku 2015).

This can be due to the fact that women are much more involved in kitchen work than men. Most of the males participate in the delivery of the already prepared food, while women are those who go bare footed during the preparation of the food, as well as those who do the washing of vegetables and fruits mainly in the kitchen (Mama and Alemu 2016).This could be due to carelessness and unhygienic habits practiced by female food handlers than male food handlers (Tefera et al. 2015).Therefore, increasing awareness about hygienic practices among food handlers is very important to enhance their health status.

In the current study, the odds of being positive for intestinal parasitic infection was associated with educational level of food handlers who educated in primary educational level (AOR: 0.055; 95% CI (0.007-0.413) which was lower than Haramaya University cafeterias, eastern Ethiopia(AOR: 2.13; 95% CI (1.24-3.67) (Marami et al. 2018). This is due to an individual's educational level greatly influences his/her health related behavior, where poor education prevents a person from realizing the importance of personal hygiene (Gezehegn et al. 2017; Marami et al. 2018). Thus, only education without practicing knowledge may not prevent his/her health from health risk factors.

Another risk factor which had statistically significant association with intestinal parasites was finger nail status. Higher intestinal parasites rate was found in those food handlers that had not trimmed fingernails than those that had trimmed fingernails (AOR: 0.330; 95 % CI (0.113-0.965).

This was lower than study conducted among food handlers in Arabaminch University in which (AOR: 2.193; 95 % CI (1.293-1.990) (Mama and Alemu 2016)and Haramaya University in which (AOR: 3.31; 95 % CI (1.99-5.49) (Marami et al. 2018). Untrimmed fingernails could serve as a vehicle for transport of organisms from the source to the food due to the area beneath a fingernail harbors most organisms and is difficult to clean (Besufikad Belachew 2017; El-Nadi et al. 2017). Thus, lack of personal hygiene might increase the probability of exposure to intestinal helminthes infection.

Hand washing with soap and water after toilet (AOR: 0.332; 95% CI (0.125-0.884) had an association with presence of intestinal parasites. Food handlers who washed their hands with soap and water was higher than Mekele University(AOR: 0.15; 95% CI (0.06-0.38) (Nigusse and

Kumie 2012) and lower than (AOR: 2.43; 95% CI (1.22-4.86) Haramaya University (Marami et al. 2018).

Level of education, nature of the working environment, limited or no sanitary surveillance made by the responsible body and availability of facilities used for hand washing practice could explain this discrepancy (Besufikad Belachew 2017). Inadequate hand washing among food handlers is a common practice that contributes to food borne diseases. Improvement of food workers' hand washing practice is, therefore, crucial to reducing the incidence of food borne illnesses.

Hence, in this study multivariate logistic regression model indicated that sex, education, untrimmed finger nail and infrequent hand washing with soap after toilet were identified as determinant factors for food handlers being infected by intestinal parasites.

These findings were consistent with the results of a study conducted among food handlers in Haramaya University, eastern Ethiopia (Marami et al. 2018) with hand washing with soap and water and finger nail ; sex with Addis Ababa University (Aklilu et al. 2015); education with Aksum town ,northern Ethiopia (Gezehegn et al. 2017). But no statistically significant difference for sex and education in a study conducted at Arbaminch University (Mama and Alemu 2016), educational status, sex and finger nail in a study conducted at Mekele University (Nigusse and Kumie 2012).

Limitation of the study

- Entamoeba histolytica and dispar cysts were not differentiated.
- Specific methods such as the adhesive scotch tape for *E. vermicularis*, Harada Moori's filter Paper for *S. stercoralis* and for hookworm infections were not done.

7 CONCLUSION AND RECOMMENDATION

7.1 Conclusion

The present studies revealed that prevalence of intestinal parasites was moderate (29.7%) of the food handlers working at Wachemo University students' cafeteria, Ethiopia were positive for different intestinal parasites. The most prevalent parasite was *A. lumbercoids* followed by Hookworms. The results of this study indicated that helminthic infections were more predominant than protozoan infections. Since most of the intestinal parasites are transmitted by the feco-oral route, food handlers could be an important source of infection to the general population. But sex, education, untrimmed finger nail and infrequent hand washing with soap after toilet were found to be associated with risk of infection with intestinal parasites and they were preventive.

7.2 Recommendations

Based on the result of the study following are recommended

For food handlers:-

- They were recommended to improve their hygiene practice.
- Those who were infected should get treatment and follow their medical status and those who were not infected should improve their hygiene practice.

For FMoH:-

- As per WHO guideline recommends periodic drug treatment (de-worming) should be given once a year when the prevalence of STH infections in the study population is over 20%.

For WU:-

- They should continuously supervise the food handlers and establish personal hygiene rule and posted it on easily visible site.
- They were encouraged to conduct regular evaluation of food handlers for their health status, including screening for the prevalence of intestinal parasites before hire them as food handlers in the cafeteria.
- Health education program should be strengthened on mode of transmission of intestinal parasites and focusing on Water sanitation and Hygiene (WaSH) practice mostly on female food handlers is pertinent to prevent acquisition of, re-infestation with and spread of intestinal parasites infections.
- Should strongly continue periodic inspection for food handlers and conduct epidemiological surveillance through quarterly routine parasitological tests and treatment of the infected cases.

For Researcher:-

- Researchers will recommended to conduct bacteriological analysis taking samples from food handlers and the environment around the cafeteria including utensils, tables, vegetables and related food stuffs.
- Further studies should be conducted by researchers to indicate the impact of food borne disease arise from carrier food handlers.

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ANNEXES

Annex I: Information Sheet

Title of the research: A cross-sectional study of prevalence of intestinal parasites and associated factors among food handlers in students' cafeteria of Wachemo University, Southern Ethiopia.

Name of Principal Investigator: Wondimagegn Desalegn Handiso

Name of the Organization: Jimma University College of Public Health Department of Environmental Health Sciences and Technology.

Purpose of the study: The aim of this study is to determine prevalence of intestinal parasites and associated risk factors among food handlers in students' cafeteria in Wachemo University. If you are willing to participate in this study you are expected to answer some questions and give stool specimens.

Procedures: In order to undertake the aforementioned study, some questions related with the topic and stool sample are taken for laboratory investigation. Permission were processed from the Jimma University and Wachemo University. Hence, you are expected to give required samples and information related with the study.

Risks associated with the study: There was no risk associated with participating in this study. You never waste time except the time required to give some information and specimen.

Benefit of the study: Study results are able to create awareness among policy makers to Strengthen/integrate existing programs that take actions on STHs. Based on laboratory result the Participants get treatments.

Confidentiality of your information: The results of the laboratory findings were kept confidential and could only be accessed by the researcher and responsible body. There was no personal information to be attached to your data.

Right to Refusal or Withdraw: Participant has the full right to refuse from participating and to withdraw at any step in this research. If you have any question you may contact the following individuals.

Based on the above information I agree to participate in the research

Signature: _____ Date: _____

Name of Data collector _____ Signature _____

Remember; if you have any question you can ask the principal investigator

Wondimagegn Desalegn Tel +251910194386 Email: - wondedslgn2000@gmail.com

Annex II: Consent Form

Greeting!

Explanation on procedures and condition of the agreement

I am from Jimma University Institute of Health, Department of Environmental Health Sciences and Technology. I am here to study about problem of intestinal parasites. The overall objective of this study was to assess magnitude of intestinal parasites and associated factors among food handlers. The information generated from this study provides the current status of intestinal parasites and associated factors in the study area. The investigation involves collection of stool for parasitological examination and interview through structured questionnaire for associated risk factors.

If the investigation was confirmed for intestinal parasite you have get treatment. I assure you the confidentially of all collected information in the questionnaire and laboratory investigation. If you have any questions regarding the purpose of the study you have the right to ask question and get clarification. It was your right to withdraw from this study if you are not interested to participate in this study. Finally, if you have understood the explanation very well, I am asking you kindly to participate in this study and put your signature as shown below.

Name of participant _____ Signature _____ Date _____

Name of data collector_____ Signature _____ Date _____

Name of principal investigator_____ Signature _____ Date_____

Annex III: Questionnaire

Part I: Introduction and consent:

Hello. My name is _____. I am working with the research team of Jimma University. We were conducting a research about intestinal parasites and associated factors. I was very much appreciating your participation in this study. The information I collect help the government to improve the health status of food handlers in food and drinking establishments. You were selected for the study. As part of the study, I would first like to ask some questions about your demographic, personal, knowledge and environmental characteristics. The questions usually take about 15 to 20 minutes. All of the answers you give were confidential and not be shared with anyone other than members of our survey team. You don't have to be in the survey, but we hope you were agreeing to answer the questions since your views are important. If I ask you any question you don't want to answer, just let me know and I was going to the next question or you can stop the interview at any time.

In case you need more information about the study, you may contact the person listed on this informed consent form.

Give information and consent sheet with contact information

Do you have any questions?

May I begin the interview now?

1. If yes, proceed to the next page,
2. If no pass to the next participant

Name of interviewer _____ signature _____

Date of interview _____ time started _____ time finished_____

Supervisors name _____ signature _____

T**hank you very much for your willingness!**

Eligibility questionnaire

1	In the last 2 to 3 weeks, did you have any of the following sign and symptoms? diarrhea (bloody or not), fever abdominal pain	
2	Are you taking antibiotic/antihelementhic drug?	1. Yes 2. No
3	If yes for the above question, when?	----------

Questionnaire for the eligible

Code ----------

Part II Participants Socio demographic/economic data			
S/N	Questions	Response options	Remarks
1.	sex	1 =Male 2=Female	
2.	Age (in years)	1= 21-30 2= 31-40 3=>40	
3.	Educational level	1=1-4 2=5-8 3=9-10 4=11-12 Grade 5=>12 Grade	

4.	Marital status	1= Single 2= Married 3= Divorced 4= Widowed	
5.	Income per month	----------	
6.	What is your job responsibility?	1= Waiter 2=Cooker 3=Equipment Collector 4=Dish Washer 5=Other/Specify/----------	
7.	Do you have training on food preparation and handling	1=Yes 2=No	
8.	If "yes" to Q 7 above , Do you have certificate	1=Yes 2=No	
9.	How about your term of service?	1= Regular/Permanent 2= Contract	
Part III Personal hygiene practice of food handlers			
10.	Finger nail status	1= Trimmed 2=Untrimmed	
11.	Do you use common knife for cutting raw flesh food and other food?	1=Yes 2=No	
12.	Do wearing clean aprons when preparing food	1=Yes 2=No	

13.	Do wearing of hair cap	1=Yes 2=No	
14.	Do cover mouth with tissue paper when coughing or sneezing accidentally during food Preparation	1=Yes 2=No	
15.	Do you have shoe wearing habit	1=Yes 2=No	
16.	Do you report when sick for your manger	1=Yes 2=No	
17.	How often do you wash hands after using the toilet?	1=Always 2=Sometimes	
18.	Do you wash hands before taking any food?	1=Yes 2=No	
19.	If yes to Q 18 to above, do you wash your hands with soap?	1=Yes 2=No	
20.	If answer is yes for Q18 above, do you wash your hands with water only?	1=Yes 2=No	
21.	Do you wash fruits and vegetables before eating?	1=Yes 2=No	
22.	Do you follow your medical check ups	1=Yes 2=No	
23.	Do you wash your cloths in the river?	1=Yes 2=No	
Part IV Environmental related factors			

24.	Where does your family get water for domestic use?	1=Stream/River 2=Tap Water 3=Borehole 4=Rainy Water	
25.	Do you carry out farming activities like weeding and digging on the farms?	1=Yes 2=No	
26.	Do you have toilet facility	1=Yes 2=No	
27.	If your answer for Q 26 is yes above, what type of toilets do you use while at home?	1=Pit Latrine 2=Water flush	
28.	Do you have separate hand washing facility?	1=Yes 2=No	
29.	Availability of shower facility	1=Yes 2=No	
30.	Where do you mostly obtain your fruits and vegetables?	1=Farm 2=Market 3= Farm And Market	
Part V Knowledge related questions about food handlers			
31.	Have you ever heard about intestinal parasites?	1=Yes 2=No	
32.	Do you know the sources of intestinal parasites?	1=Yes 2=No	
33.	If your answer is yes to Q 32 above, what are the sources of intestinal parasites?	1=Contaminated Food 2=Contaminated Water	

		3=Health Professionals 4=Other/Specify/_____	
34.	Can intestinal parasites be transmitted?	1=Yes 2=No	
35.	If yes to Q 34 above, how could it be transmitted?	1=Direct Contact 2=Faecal Contamination 3=Sexual Contact 4=Other/Specify_____	
36.	Can some intestinal parasites be transmitted during bare foot movement?	1=Yes 2=No	
37.	If your answer is yes to Q 36 above, which parasite? Specify	Specify_____	
38.	Can some intestinal parasites be transmitted during swimming?	1=Yes 2=No	
39.	If your answer is yes which parasite?	Specify_____	
40.	Can intestinal parasites preventable?	1=Yes 2=No	
41.	If your answer is yes, do you know how intestinal parasites could be prevented?	1=Personal Hygiene 2=Drinking Alcohol 3=Environmental Sanitation 4=Food Hygiene 5=Eating Cooked Food/Vegetable 6=Other/Specify_____	
42.	Do they have treatment?	1=Yes 2=No	

43	Do you think intestinal parasites cause fata disease?	1=Yes 2=No	

Part V Participants Laboratory Data		**Stool**	**Ova**
Parasites isolated from Stool/Ova	*A.lumbrcoid*		
	T. trichuria		
	E.hiistolica/dispar		
	S.stercolaris		
	Hook worm species		
	G.lamblia		
	H.nana		
	Taena species		
	Mixed		
	Others/specify_____		

Annex IV: - Laboratory Result Record Format

<table>
<tr><td colspan="2">Jimma University Institute of Health Faculty of Public Health Department of Environmental Health Sciences and Technology</td></tr>
<tr><td colspan="2">A cross-sectional study of the carrier status of intestinal parasites among food handlers in students cafeteria of Wachemo University, SNNPR Ethiopia, 2019.</td></tr>
<tr><td colspan="2">Laboratory request form.</td></tr>
<tr><td>Code/labeled number.
Age _____
Sex F ☐ M ☐</td><td>Time of sample collection_____/_____
Date of sample collection _____/_____</td></tr>
<tr><td>Physical examination of stool</td><td>Formed ☐ Semi formed ☐ Bloody ☐
Watery ☐ Other/specify ☐</td></tr>
<tr><td colspan="2">Laboratory Result/s
A. Intestinal parasites seen B. No intestinal parasites seen</td></tr>
<tr><td rowspan="2">Direct wet mount microscopic examinations</td><td>Intestinal Parasites Identified</td></tr>
<tr><td>A.lumbrcoid ☐
T. trichuria ☐
E.hiistolica/dispar ☐
S.stercolaris ☐
Hook worm species ☐
G.lamblia ☐
H.nana ☐
Taena species ☐
Mixe ☐
Other/specify _____</td></tr>
<tr><td></td><td>A.lumbrcoid ☐
T. trichuria ☐</td></tr>
</table>

☐

Formalin Ether (ethylacetate) method	*E.hiistolica/dispar* *S.stercolaris* ☐ *Hook worm species* ☐ *G.lambli* ☐ *H.nana* ☐ *Taena species* ☐ *Mixed* ☐ *Other/specify* _____
Laboratory technicians/technologists name _____ Signature _____date _____/_____/_____	

Annex V: Observational checklists

S/n	Questions	Response options	Remarks
1.	Does the workers wear gown?	1=Yes 2=No	
2.	Does the worker wear hair cover?	1=Yes 2=No	
3.	Are finger nails trimmed and clean	1=Yes 2=No	
4.	Does the workers gown clean	1=Yes 2=No	
5.	Discharging from nose, eye, ear and cough during visit	1= Observed 2= Not observed	
6.	Wear of any jewelry or ring at time of visit	1= Observed 2= Not observed	
7.	Does nail paint observed?	1=Yes 2=No	
8.	Do have regular medical check-up? see the evidence	1=Yes 2=No	
9.	Are you excluded from food preparation or serving when you are sick?	1=Yes 2=No	
10.	Utensils and equipment are cleaned and sanitized between uses	1=Yes 2=No	
11.	Are the fixtures, fittings and equipment maintained in a clean condition?	1=Yes 2=No	

Annex VI: Eyyexxi Dabachcha /Consent Form Hadiyegna Version /

Xumma!

Ka sorobim bikkina yooki duhaa itanchcha cakishsha

Anni Jimmi Universitee fayyommi shoggi lossancho.Anni ka univresitinne goddaphi henshish Bikkinna Sorrobimma isommanne. Kasorrobimmik horror woshi lossanʿiihurabbaxi minnine Baxxo baxannekka godaphi henshish bikkinna ka hensheshinne amaxxammo luwwuwibikkinna sorribimma. Ku sorrobima madokkoki henshesha jabbo amadiso fochii llechuww ixininisiminna cirroaʿimmakajabbine amaxamoluwwibikkina guddakko xammichuwwine xammimma. Laborrtoorree dabachissinne ku jabbi sidamulasse qararee sidakkammo.Anni kinisse wixaʿummil Uwwa hundamme ayyename kurrunnoi ibeyyonne amadommanne.

Ka anni issummo sorobimmanne agubbe luwwi xamichiyo lasse xammitakkenna xantakammo.Kasooribbimannee xximmina hasi beelasee uullisima xanatittoo.Lassanchonne ku yummi luwwi hundimm adanamissaagaʿ kollase ki eyyittomma worronne gudukkibeyyone Summa kitabinne marree dissimine laʿisse.

Gallaxxommo!

Xamamachi summa _____ marree _____ ayyammo _____

Xammanchi summa _____ marree _____ ayyammo _____

Sorrobboawannisachi summa _____ marree _____ ayyammo _____

Annex VII: Xammichuwwa /Questionnaires Hadiyegna version/

Annanximareʽe ----------

Qoxxo I Ani anichi gaqi halato labo duhuwwa			
Xigo	Xamicha	Dabacha	Sawwitte
1.	Albachcha	1 = Goncho 2=Menticho	
2.	Uumurr hinchinne	1= 21-30 2= 31-40 3=>40	
3.	Lossanni gabbala	1=1-4 2=5-8 3=9-10 4=11-12 5=>12	
4.	Min isaqanchi duhha	1= Mullateʽee 2=Manchiyohanne 3= Manichi lehakohanne 4= Manchi anani ihamohane	
5.	Agannisixoʽimeeʽoo	----------	
6.	Baxi shardi maricho	1= Dananimma 2=Sarimma	

		3=Mutta wixaiʾmma 4=Mutta anshimma 5=Mullekee----------	
7.	Hurabata gudisimanne boradisha masitahinne	1=Oyya 2=Masumoyyo	
8.	Xamich“7” dabachi oyya ihulasi boradish woraqati beeʾanne	1=Oyya 2=Beeʾe	
9.	Baxi duhaʾihinkidette	1= Adil baxanchotte 2= Amanominatte	
Qoxxo II Hurabaxi Baxanni gaqi mucuromi bikkinna			
10.	Angi xuranqi duha	1= Murramma 2=Muramubeʾanne	
11.	Hurabatta ananni billawaa awaxitta muttuhonne	1=Oyya 2=Awaxomoyo	
12.	Mucuri baxi edeecha edollohonne	1=Oyya 2=Edoʾmoyyo	
13.	Horolli qobbee heʾanne	1=Oyya 2=Beeʾe	
14.	Kuxisoammanesummesannnemucuriluwwinneef assohonne	1=Oyya 2=Iffisomoyyo	

15.	Kobbeˈeawaximmihalatiyoˈonne	1=Oyya 2=Beeˈe	
16.	Dishi yitakkmiamanne awanasachinna kuttohonne	1=Oyya 2=Kurromibeˈanne	
17.	Ciili mini lasganne hinkanni amanina anaga ashaqitakamo	1=Hundi amaneem 2= Higahiga 3=Mat imatiamanne 4=Horriyyemme	
18.	Hinki hurriba tatami amadim millagea naga anshitohonne	1=Oyya 2=Anishaqobeˈanne	
19.	Xamich“18” dabachi oyya ihulasi anaga samuninne wo’nne anashaqitohonne	1=Oyya 2=Anishaqo beˈanne	
20.	Xamich“18” dabachi oyya ihulasi anaga samuninne wo’nne anashaqitohonne	1=Oyya 2=Anishaqobeˈanne	
21.	Mishuwa itimmi illage anshitohhone	1=Oyya 2=Anishaqo beˈanne	
22.	Goddab ihenshishbikinna qararemasitohonne	1=Oyya 2=Masumoyyo	
23.	Dajenne edecha anshitohonne	1=Oyya 2=Anshomoyyo	

Qoxoʹi III Hegeqqi Bikkina			
24.	Mini awadinna woʹo hanisidakammo	1=Daji 2=Bombi 3=Barri 4=Xenni	
25.	Abulli baxo baxohonne	1=Oyya 2=Baxomoyyo	
26.	Cilli mini yoʹonne	1=Oyya 2=Beeʹe	
27.	Xamich “26” dabachi oyya ihulasi hinkido cill mini yokko	1=Haqqinne baxamokoki 2=Woʹimallayine baxokoki	
28.	Ananni anaga anshaqakkami woʹiyoʹonne	1=Oyya 2=Beeʹe	
29.	Oracho anshaqim min yoʹonne	1=Oyya 2=Beeʹe	
30.	Lobakati amanne mishuwa hani sidoto	1= Abulli 2=Meeri	
Qoxxoʹi IV Qorrommanne Lachi Bill Xamicha			
31.	Heneshesh bikkina macessa laqohonne?	1=Oyya 2=Laʹomoyyo	

32.	Henshesh hani warode elaqohonne?	1=Oyya 2=Laˈomoyyo	
33.	Xamich “32” dabachi oyya ihulasi hashish jab waroo beyyuwi hanihano?	1=Jorihuribatta 2=Joriwoˈo 3=Fayoˈomibaxanno 4=Mulleki _____	
34.	Godaphi henshesh jabi matimatonne higgone?	1=Oyya 2=Higo beeˈanne	
35.	Xamich “34” dabachi oyya ihulasi hinkic higgena xanokko	1= Nakachinne 2=Cirinnenakachine 3=Shaˈixiedanachine 4=Mulleke _____	
36.	Mati mati henshish jabawi lexaˈi lokkine takimmine higonne	1=Oyya 2=Higo beeˈanne	
37.	Xamich “36” dabachi oyya ihulasi mayyaka mihenshish jab ihigokko	kurre_____	
38.	Mati mati henshish jabawi lexaˈi lokkine takimmine higonne	1=Oyya 2=Higo beeˈanne	
39.	Dabachi oyya ihulasi mayakami henshesh jabi	Kure _____	
40.	Ka henshish jabo hollamenna xannomone	1=Oyya 2=Xannomoyyo	

41.	Dabachi oyya ihulasi hinkido gogine hollamenna xannomo	1=Gaqi mucurommine 2=Dimbiso aga agiminne 3=Hegeqi mucurommine 4=Hurribaxi mucurommine 5=Sarami hurribata itimmine 6=Mulleke_____	
42.	Qarari yohannenne	1=Oyya 2=Beʾanne	
43.	Shenna xanohonne	1=Oyya 2=Xannoyo	

Annex VIII: SOP for Direct wet mount stool Specimens examinations

Procedure for Direct Wet Mount preparation

1. Using pencil or marker label the slide by food handlers' code number.

2. Place a drop of fresh physiological saline on one end of a slide and a drop of iodine on the other end.

3. To avoid contaminating the fingers and stage of the microscope, do not use too large a drop of saline or iodine.

4. Using a wire loop or piece of stick mix a small amount of specimen, about 2mg, (matchstick head amount) with the saline and a similar amount with the iodine. Make smooth thin preparations. Cover each preparation with a cover glass.

Note: Sample from different areas in and on the specimen or preferably mix the faeces before sampling to distribute evenly any parasites in the specimen. Do not use too much specimen otherwise the preparations will be too thick, making it difficult to detect and identify parasites.

5. Examine systematically the entire saline preparation for larvae, ciliates, helminthes eggs, cysts, and oocysts. Use the 10x objective with the condenser iris closed sufficiently to give good contrast. Use the 40x objective to assist in the detection and identification of eggs, cysts, and oocysts.

6. Always examine several microscope fields with this objective before reporting "No parasites found."

7. Use the iodine preparation to assist in the identification of cysts

8. Record the findings.

9. Report by specifying the parasite name and the stage of development (egg, larvae, trophozoite and cyst in the entire saline preparation.

Annex IX: SOP for Formol Ether Concentration Technique

Materials

- Formol water, 10% v/v.
- Prepare by mixing 50 ml of strong formaldehyde solution with 450 ml of distilled or filtered rain water.
- Diethyl ether or ethyl acetate.
- Sieve (strainer) with small holes, preferably 400–450_m in size.
- The small inexpensive nylon tea or coffee strainer available in most countries is suitable (can be used many times and does not corrode like metal sieves).

Procedures

1. Label screw caped test tubes with unique specimen number.
2. Add about 4ml of formol water solution (10% v/v) into each labeled test tubes.
3. Using applicator stick, emulsify an estimated 1g (pea-size) of faeces into each tube.
4. Note: Include in the sample, faeces from the surface and several places in the specimen.
5. Add a further 3–4 ml of 10% v/v formol water and cap the bottle.
6. Mix well by shaking.
7. Sieve the emulsified faeces and collect the sieved suspension in a beaker.
8. Transfer the suspension to a conical (centrifuge) tube.
9. Add 3–4 ml of diethyl ether or ethyl acetate.

Caution: follow universal safety precautions

10. Stopper the tube and mix for 1 minute.
11. Centrifuge immediately at 750-1000g for 1 minute.

12. After centrifuging, the parasites will have sediment to the bottom of the tube and the fecal debris will have collected in a layer between the ether and formol water.

13. Using a stick or the stem of a plastic bulb pipette loosen the layer of fecal debris from the side of the tube and invert the tube to discard the ether, fecal debris, and formol l water. The sediment will remain.

14. Return the tube to its upright position and allow the fluid from the side of the tube to drain to the bottom.

15. Tap the bottom of the tube to re-suspend and mix the sediment.

16. Transfer the sediment to a slide, and cover with a cover glass.

17. Examine the preparation microscopically using the 10x objective with the condenser iris closed sufficiently to give good contrast.

18. Use the 40x objective to examine small cysts and eggs.

19. To assist in the identification of cysts, run a small drop of iodine under the cover glass.

(Tamiru et al. 2015)

Annex X: Ethical Clearance Letter

Ref.No. IHRPGD/ 713/2019

Date: 26/02/ 2019

Institutional Review Board (IRB)
Institute of Health
Jimma University
Tel: +251471120945
E-mail: zeleke.mekonnen@ju.edu.et

To: Wondimagegn Desalegn Handoso

Subject: Ethical approval of research protocol

The IRB of institute of health has reviewed your research project entitled:

"Prevalence of intestinal parasites and associated risk factors among food handlers in Wachemo University students' cafeteria"

This is to notify that this research protocol as presented to the IRB meets the ethical and scientific standards outlined in national and international guidelines. Hence, we are pleased to inform you that your protocol is ethically cleared.

We strongly recommended that any significant deviation from the methodological details indicated in the approved protocol must be communicated to the IRB before they are implemented.

With regards!

Zeleke Mekonnen (PhD)
Associate Professor, Health
Research and Postgraduate
Director

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Declaration

I declare that this research paper is my original work and has not been presented for a degree in any other university and that all sources of materials used for the research paper have been correctly acknowledged.

Name: Wondimagegn Desalegn Handiso

Signature: ---------- Date ----------

This research paper has been submitted for examination with my approval as an advisor:

1. Name ---------- -Signature ----------Date----------
2. Name ----------Signature ----------Date----------
3. Name ----------Signature ----------Date----------

The result of this thesis has been submitted for the department of environmental health Sciences and technology with my approval as a University examiner.

Internal examiner

Name ----------Signature ----------Date of approval ----------

External examiner

Name ----------Signature ----------Date of approval ----------