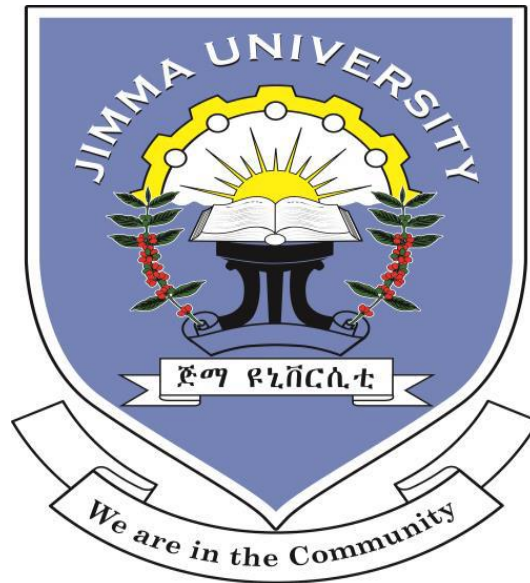


**INTESTINAL PARASITIC AND BACTERIAL CONTAMINATION OF
FRESH/RAW VEGETABLES AND FRUITS SOLD IN OPEN-AIRED
MARKETS IN PERI-URBAN KEBELES OF JIMMA TOWN,
SOUTHWEST ETHIOPIA**



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**MAY, 2022
JIMMA, ETHIOPIA**

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

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ABSTRACT

Background: *Vegetables and fruits are essential for a healthy human body and they form a major component of the human diet. Consuming fresh vegetables and fruits reduces the risk of chronic diseases. However, human infection with medically important parasitic helminthes, protozoa, and bacteria due to consumption contaminated raw vegetables and fruits coupled with lack of sanitation and hygiene have been a major concern in developing countries including Ethiopia.*

Objective: *The study was to assess intestinal parasitic and bacterial contamination of fruits and vegetables, sold in open aired markets and assess KAP of Sellers and associated factors in peri-urban Kebeles of Jimma town, Southwest Ethiopia.*

Method: *A community based cross-sectional study was conducted in three peri-urban Kebeles of Jimma town, Southwest Ethiopia. Three hundred seventy-five fruits and vegetables were purchased from 375 sellers by one to one ratio and simple random sampling technique was used to select sellers and fruits and Vegetables. Data were collected by questionnaire. For parasitic identification, the sediment was examined under a light microscope and modified Zeihl-Neelsen staining technique while bacterial contamination was identified by growing in the MacConkey agar, Mannitol salt agar and Salmonella-shigela agar culture media. Data were entered into Epi data version 4.6 and exported to SPSS version 25. Logistic regressions were performed to determine the association with a confidence interval of 95%, p-value ≤ 0.05 was considered statistically significant.*

Results: *A total of 375 fruits (n=188) and vegetables (n=187) were collected from three site peri urban kebeles of Jimma town. Of the total fruits and vegetables, 173 (46.1%) and 194 (51.7%) samples were found contaminated with at least one type of parasite and bacterium, respectively. The most frequently encountered parasitic egg/cyst was *A. lumbricoides* 46(12.3%), followed by *Strongyloides stercoralis* 32(8.5). Nine (9) species of bacteria were detected; *S. aureus* being the commonest bacterial contaminant of fruits and vegetables (77, 20.5%) followed by salmonella species (66, 17.8%). AST was performed for isolated bacteria specious and Ciprofloxacin was the most effective drug (79.9%) followed by Cefazoline (77.7%), cotrimoxazole (74.58%), ceftriaxone (66.4%), and Gentamicin (61.15%). But Highest resistance was observed in penicillin (84.4%) followed by ampicillin (81.4%).*

Conclusions and Recommendations: *Parasitic and Bacterial contaminations were high in fruits and vegetables sold at markets of peri-urban kebeles of Jimma Town. Local and regional government should regulate vendors handling practices and regular on market screening should be done. Moreover, attentionn should be given on rational use of drugs to prevent microbial resistance.*

Key wards: *Fruits, Vegetables, Parasite, Bacteria, Jimma*

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ABBREVIATIONS AND ACRYNOMS

CDC - Centers for Disease Control and Prevention

CFU - Colony Forming Unit

KAP- Knowledge, Attitude and Practice

NTDs - Neglected Tropical Diseases

WHO - World Health Organization

CHAPTER ONE

1. INTRODUCTION

1.1 Background

Vegetables and fruits are essential for a healthy human body and they form a major component of human diet. Moreover, vegetables are essential sources of energy and substantially improve food quality as rich sources of water, vitamin C, carotene, mineral elements such as iron, and vitamins including thiamine (vitamin B12), niacin and riboflavin(1). Consuming fresh vegetables and fruits reduces the risk of stroke, cardiovascular diseases, and has been shown to protect against certain types of cancer(1,2). The recent increase in awareness about the health benefits of vegetables has ensued in increased demand to consume it. Because of health benefits, World Health Organization (WHO) recommends intake of sufficient fruits and vegetables (400g) per day, usually not only decrease the risk of non-communicable diseases(NCD)(3), but also used for the prevention and mitigation of several micronutrients deficiencies. Inadequate consumption of vegetables and fruits contributes to poor health and increase the risk of chronic diseases; such as heart diseases, diabetes, cancers and obesity(4).

Most of the time, raw vegetables are eaten in a variety of ways, such as mixed greens to preserve the common flavor and warm labile nutrients, and this practice may increase the likelihood of parasite disease transmitted through food. Fruits and vegetables, especially those eaten raw and without peeling, are a public health problem because they provide a safe haven for pathogenic parasites and bacteria (5).

However, human infection with medically important parasitic helminthes and protozoa due to improper utilization of crude vegetables and fruits coupled with need of sanitation and hygiene has been a major concern in developing countries including Ethiopia. Studies have shown that parasites such as *Ascaris lumbricoides*, *Cryptosporidium* species, *Entamoeba histolytica*, *Enterobius vermicularis*, *Fasciola* species, *Giardia intestinalis*, hookworm species, *Hymenolepis* species, *Taenia* species, *Trichuris trichiura*, and *Toxocara* species can infect humans who consume contaminated, uncooked, or improperly washed vegetables and fruits(6–13).

Enteric bacterial infections are disseminated nearly all through the world and have been causing morbidity among communities where poor environment hygiene and personal hygiene are widespread. Enteric pathogens such as *E.coli*, *Salmonella* and *Shigella species* are among the greatest concerns during food related outbreaks. The increase in these food borne infections may have resulted from increased consumption of contaminated fruits and vegetables (14).

The most common bacteria identified in vegetables and fruits include *Staphylococcus aureus*, *Enterobacter species*, and *Klebsiella species*. *Escherichia coli*, *Salmonella typhi*, and *Serratia species*. *E.coli* is a common human pathogen that causes diarrhea, kidney failure, pneumonia, skin infection, respiratory disease, meningitis, and food poisoning, among other illnesses. This is more common in persons who have weakened immune systems (15).

The presence of *Staphylococcus aureus*, a pathogenic organism of public health concern, and the presence of other pathogenic and opportunistic bacteria like *Salmonella spp.* and *Klebsiella spp.* in some of the fruits and vegetables, further need to safeguard the health of the consumers by proper washing and decontamination of these fruits and vegetables which are eaten raw (16–18).

Eating raw vegetables and fruits is common practice among rural communities in Ethiopia. Such practice may raise the risk of parasite and bacterial infections in the intestine, especially if the vegetables and fruits are not washed and cooked thoroughly (19). Given the importance of fruits and vegetables, it is essential to conduct periodic assessments to determine the prevalence of parasites and pathogenic bacteria on fruits and vegetables eaten by the general public. The aim of this study is to assess intestinal parasitic and some pathogenic bacterial (*Enterobacteriaceae* like *E.coli spp*, *salmonella spp*, *Shigela spp*, *klebsiella spp* and pathogenic gram positive cocci like *S. auerus*) contamination of vegetables and fruits sold in open-aired markets and to assess KAP of sellers/vendors about contamination of fruits and vegetables by intestinal parasites and bacteria, that sold in open aired markets in peri-urban Kebeles of Jimma town, Southwest Ethiopia.

1.2. Statement of the Problem

1.2.1. Intestinal parasitic Contamination

As WHO reported in 2020, intestinal parasitic infections remain the global health problem, which more affect the low-income countries with poor sanitation practice. Nowadays worldwide, the Soil transmitted helminthes infection affects around 1.5 billion population (20). Intestinal protozoa (particularly *E.histolytica* and *G.lamblia*) have infected over half a million individuals worldwide, resulting in over 10,000 deaths in settings where sanitation and hygiene are inadequate. Contaminated food and water diseases are a primary source of morbidity and mortality, and they continue to constitute a serious threat to public health. The consumption of contaminated raw vegetables and fruits is a major epidemiological factor in the spread of parasitic foodborne diseases(21–23).

Several studies in different partsof the world shown that contaminated vegetables can be cause for transmission of protozoa cysts, and oocytes, such as *Giardia*, *Entaemeoba*, *cryptospora* and *helminthes* eggs and hatchlings of *Hymenolopis*, *Teania*, *Fascoila*, *Ascaris*, *Strongoloides*, and Hookworms(11,24,25)

In Africa countries, the transmission of intestinal parasitic contamination has been considered to extend due to the utilize of untreated human and animal dung as fertilizer and untreated waste water for irrigation during cultivating by nearby agriculturists, which serve as source of parasitic contaminations and the main contributing factors in the pre harvesting phase (26,27). A study has been reported from Nigeria showed that on parasitic infection, contamination of fruits and vegetables are due to geohelminths' ova and protozoan cyst (28).

Like other developing countries, Ethiopia is a country with a poor sanitation and hygiene which makes it vulnerable to intestinal parasitic and bacterial contaminations. The studies in Ethiopia showed that fruits and vegetables sample collected from different markets contaminated by parasites were, in Bahir Dar City, contamination rate of fruits alone was 25.6% and Vegetables accounted for 49.1% of the total, in Tarcha town, the overall contamination rate was 42.6% and the overall contamination rate in Jimma Town was 57.8%(10,13,29).

1.2.2. Bacterial contamination

According to CDC (Center for diseases control and prevention), in recent years, there has been an increase in the rate of bacterial contamination of fresh fruits and vegetables. Thus, these public concerns are well-founded, as many foodborne disease outbreaks have been linked to the ingestion of fresh vegetables infected with bacteria such as *Listeria monocytogenes*, *Escherichia coli* O157:H7, and *Salmonella* spp (30,31).

A study from India reported that occurrence of *E.coli* in fruits and vegetables sold at local markets. The food items which directly consumed or used in salads or similar preparations, thus, hazardous to public health. The incidence of bacterial inhabitants on vegetables and fruits is known as a source of potential health danger to human (32). Some studies in Ghana suggest that vegetables sold in Groceries and in the markets are contaminated with microorganisms such as Bacteria and parasites (30,33).

In Ethiopia, the chance related with presentation of outbreaks of foodborne illness like sullied vegetables primarily due to lack of awareness on sanitation particularly amid the blustery season. The unsafe Ethiopian custom of eating raw food without pretreatment with insufficient heat or detergents, which is practiced across the country. This is because vegetables and fruits which are sold in the market can bring other side health risks since it mostly acts as a reservoir for many microorganisms (34,35). Reduction in the incidence of food-borne illnesses is strongly influenced by the knowledge, attitudes and practice of food-handlers towards the implementation of food safety plans. Thus, there is a strong linkage between positive behavior, attitudes and education of food-handlers in maintaining safe food handling practices (1,1). The knowledge, attitudes and practices (KAP) of food-handlers have been reported in studies from different countries around world including our country Ethiopia (2–5, 5–8). However previous studies in Ethiopia were concentrated on food establishment workers and to the best of our knowledge there is no previous study in Ethiopia assessing KAP of fruit and vegetable vendors. Fruits and vegetables plays a role as a carrier and reservoir of antibiotic resistant bacteria (9,10).

Developing countries like Ethiopia were new fruits and vegetables are for the most part delivered by farmers and sold by vendors who have small or no information of food borne illness will proceed to confront the challenge of defilement. The previous attempt was to determine the level of bacterial contamination of selected vegetables and antimicrobial property(36) and to determine the level of parasitic contamination of selected fruits and

vegetables and associated factors(10). So, both studies showed that the importance of raw fruits and vegetables as the potential source of transmission for intestinal parasites and bacteria to humans(10). Therefore, this study, was attempt not only to determine the extents of raw fruits and vegetables contamination with parasites that was transmitted to human and associated factors and to determine the level of bacterial contamination in fruits and vegetables that sold in open aired markets and antimicrobial property but also to assess Knowledge, attitude and practice of vendors/sellers in peri-urban Kebeles of Jimma town, Jimma, southwest Ethiopia.

1.3 Significance of the study

Ethiopia is a country with a low Sanitation and hygiene, which makes it vulnerable to intestinal parasites and bacterial infections. Even though number of researches was conducted on contamination of raw fruits and vegetables with medically important microorganism such as parasites and bacteria, focused on urban kebeles but not on peri- urban kebeles. Peri-urban areas are those which are characterized by a mix of rural and urban characteristics. So, to our knowledge there's no published document on contamination of raw fruits and vegetables with medically important microorganism such as parasites and bacteria in the peri-urban kebeles of Jimma town. And also, previous studies didn't assess the KAP (knowledge, attitude and practice) of sellers/vendors about Intestinal parasites and bacterial contamination of fruits and vegetables.

This study will help to provide useful information to the role of concerned body in the control of parasitic and bacterial contamination in these vegetables and fruits

It will be used as a base line for those who are interested in carrying out further study

Also, it will help to aware vendors/people about health risks that possible associated with consuming unhygienic fruits and vegetables that sold in open aired markets.

CHAPTER TWO

2. LITERATURE REVIEW

2.1. Intestinal Parasitic contamination

Intestinal parasites infections remain the major health problem worldwide especially in low-income countries. Globally the greatest numbers intestinal parasites infection occurs in Sub-Saharan Africa, the Americas and Asia. More than 100 countries, including Ethiopia, are endemic for soil-transmitted helminthes infections (37). In Ethiopia, 21.6 million people live in areas where schistosomes and soil-transmitted helminthes are endemic, and 44.6 million people live in areas where schistosomes and soil-transmitted helminthes are endemic, respectively. The protozoan infection is also spreading across the country (38).

The Study Performed in Hue city, Vietnam, showed that from vegetables types collected from three markets, all vegetable samples were contaminated with parasites. *Fasciola* (eggs, 83.33 %), *Ascaris* (eggs, 85.19 %), and *Trichuris* (eggs, 85.19 %) were the parasites most usually found (eggs, 64.81 %). *Clonorchis sinensis* eggs were also found in 16.67 %. On the test samples, oocysts of certain protozoa such as *Cryptosporidium*, *Isospora*, and *Cyclospora* were also identified. *Cryptosporidium* contamination levels ranged from 22.22 % to 66.67 % on 12 different types of vegetables tested. *Isospora* and *Cyclospora* were found in a small number of samples. Watercress, basil, and lettuce were the most parasite-infested of the twelve varieties of vegetables examined (39).

Studies conducted in Nigeria showed that, contamination of vegetables by intestinal parasites was (11.6%). Cabbage had 11 (22%), the highest number of samples contaminated with intestinal parasites, followed by Carrot 7(14.0%) and Lettuce 6(12.0%). Three nematode stages recovered from the samples included eggs of *Ascaris lumbricoides*, larvae of *Strongyloides stercoralis* and rhabditiform larvae of hookworm (40).

Other study conducted in, Nigeria, results revealed fruits and vegetables were contaminated with parasite cysts, eggs or larva. From total samples studied (41.67%) samples were found to be positive for parasites during the study. Parasites found included *Ascaris lumbricoides*, *Trichuris trichiura*, Hookworm, *Strongyloides stercoralis*, *Schistosoma spp*, *Fasciola spp*, *Entamoeba coli*, *Giardia lamblia*, *Entamoeba histolytica*, *Taenia spp* and *Entamoeba*

hartmanni. Results from the study showed highest parasitic contamination of fruits and vegetables with *A. lumbricoides*, 11 (10.19%), while *S. stercoraris*, *Schistosoma spp*, *G. lamblia* and *E. hartmanni* least contaminated the fruits and vegetables 1 (0.93%) each (41).

The study performed in Benha, Egypt, showed a considerably high level of contamination of green vegetables involving intestinal parasites (29.6%), among the vegetables, lettuce is the most contaminated.(45.5%), followed by watercress (41.3%), parsley (34.3%), green onion (16.5%), as well as leek, which is the least contaminated (10.7%) and intestinal parasitic were detected in that study, *Giardia lamblia* cysts were the most prevalent parasitic stage contaminating green vegetables (8.8%), *Entamoeba spp.* cyst (6.8%), *Enterobius vermicularis* (4.9%) , *Hymenolepis nana* 2.8% and *Ascaris lumbricoides* (0.6%)(11).

The Study Performed in Tarcha town, Ethiopia, showed that from fruit and vegetable samples examined, 42.6% were found contaminated with at least one type of parasite. *A. lumbricoides* ova, *Toxocara spp.*, *Hymenolepis nana*, and *Hymenolepis diminuta* oocysts, and cysts of *G. intestinalis* and *E. histolytica/dispar* were among the parasites identified. The most common parasite identified was *A. lumbricoides* (16.7%), followed by *Toxocara* (13.7%), *Hymenolepis nana* (11.9%), *H. diminuta* (10.7%), *E. histolytica/dispar* (10.4%), *G. lamblia* (9.6%), and *Cystoisospora belli* (9.6%) (13).

The study conducted in Jimma Town showed that, from examined fruits and vegetables samples the results of the study showed that, 57.8% samples were identified to be contaminated with at least one type of parasite. Green pepper accounts for 53%, cabbage for 68.9%, lettuce for 55.6 %, salad for 77.8%, carrot for 62.2 %, tomato for 46.7 %, banana for 51.1 %, and mango for 46.7 %. Strongyloides like parasite (21.9%) was the parasite contamination most usually found, followed by *Toxocara Spp* (14.7%), *Cryptosporidium Spp* (12.8%), *H. nana* (8.3%), *G. lamblia* (7.5%), *A. lumbricoides* (6.7%), *E. histolytica/dispar*(5.3%), *Cyclospora spp* (5.0%), and *H. diminuta* (5.0%) (10).

2.2. Bacterial Contamination

As Study Worked at Karnataka, India, indicated that among vegetable samples analyzed from different markets (35.27%) samples showed cultural positivity. *E.coli* constituted the majority of the isolates (38.14%) closely followed by Salmonella (36.59%) and significantly Shigella sp, the causative agent of bacillary dysentery was also present in 49 (25.25%) samples. Most highly

contaminated vegetables are chilli 87.50%, dill 75.00%, capsicum 70.00%, spinach 66.67%, parsley 66.67%, peas 60.00%, cauliflower 50.00%, Potato 43.33%, Fenugreek leaves, carrots, and gongura account for 42.86 %, 42.86 %, and 41.67 %, respectively. Cucumber 34.15 %, tomato 30.26 %, onion 24.44 %, ivy gourd 38.46 %, coriander leaves 26.67 %, bitter gourd 30.00 %, drumstick 21.42 %, ladies finger 20.00 %, French beans 22.73 %, pumpkin 21.73 % are moderately contaminated vegetables, whereas zucchini 14.71 %, snake gourd 12.5 %, brinjal 7.69 %, sponge gourd 6.67 % are the least (42).

The study conducted in Hue city, Vietnam, showed that from total samples of vegetables types were collected from three markets for Evaluation of Aerobic Bacteria and *E. coli* contamination and salmonella contamination, the study results showed that all vegetable samples were contaminated with aerobic bacteria and *E. coli*. Aerobic bacteria and *E. coli* concentrations varied from 6.84 to 8.40 log CFU/g and 5.47 to 6.88 log CFU/g, respectively. In the same type of vegetable, there was a considerable difference in *E.coli* load between the three markets, particularly in celery, cilantro, and Vietnamese cilantro. Meanwhile, water spinach had the largest variation in total aerobic bacteria concentration (7.52–9.45 log CFU/g), followed by Vietnamese cilantro (6.82–7.81 log CFU/g)(39).

The study was conducted in Cape Coast in Ghana, to evaluate the microbiological quality of cabbage, lettuce and scallions sold in local markets. Bacteria were found in the vegetables used in this study. *Enterobacter spp.*, *Escherichia coli*, *Klebsiella spp.*, *Salmonella spp.*, *Serratia marcescens*, and *Staphylococcus aureus* contaminated the vegetables(30).

As the study carried out in Nigeria, the results of the microbial contamination of fruit samples collected from different fruit vendors were Examined for contamination of bacterial infection shows the result of the average microbial load of the vended fruit samples in Colony forming unit per ml (CFU ml⁻¹).Tiger nuts have the greatest average total aerobic plate count of 1.03 x10⁶, followed by watermelon (sliced), which has 1.0 x10⁶, and cucumber, which has 3.5 x 10⁵. In addition, the study shows the result of the morphological and biochemical characteristics of the microbial isolates from the ready-to-eat vended fruit samples. *Salmonella sp*, *Pseudomonas sp*, *Escherichia coli*, *Shigella sp*, and *Staphylococcus aureus* were found to be among the bacteria isolated. Except for *Staphylococcus aureus*, which is cocci in form and Gram positive, all of the bacterial isolates are rod-shaped and Gram negative. Moreover, all

bacterial isolates are catalase positive while four bacterial isolates are coagulase positive except for *Pseudomonas* sp which is coagulase negative(43).

According to the study conducted in local markets of Arba Minch town, Ethiopia showed that a total of 48.7% of the vegetable samples collected from local markets were found to be infected with bacteria. Cabbage was the most commonly infected vegetable (71.9%), followed by carrots (56.5 %).The *E. coli* was the most common bacterial contaminate of vegetables, with four different species detected (31.4 %)(7).

The study conducted in Jimma town showed that eight genera of bacterial isolates from vegetables which were identified; *Enterobacter spp.* (21.60 %), *Citrobacter spp.* (20.6 %), *Klebsiella spp.* (18.6 %), *Salmonella spp.* (11.8 %), *E. coli* (10.8 %), *Proteus spp.* (9.8 %), *Staphylococcus spp.* (4.9 %), and *Pseudomonas aeruginosa* (4.9 %) were the most common bacteria(2%)(91).

Generally, raw fruits and vegetables are known to have the potential for harboring a wide range of microorganisms and causing several outbreaks. According different studies mention above, the results of the studies showed us Foodborne bacterial pathogens commonly detected in fresh vegetables and fruits were different toxin producing *E.coli strains*, *Staphylococcus aureus enterotoxin*, *Salmonella species*, *Shigella species*, *Bacillus species*, *Campylobacter species*, *Listeria monocytogenes*, *Klebsiella spp.*, *Pseudomonas aeruginosa*, *Proteus spp.*, etc and intestinal parasites such as *Ascaris lumbricoides*, *Cryptosporidium species*, *Entamoeba histolytica*, *Enterobius vermicularis*, *Fasciola species*, *Giardia intestinalis*, hookworm, *Hymenolepis species*, *Taenia species*. *Trichuris trichiura* and *Toxocara species* can infect humans who consume contaminated, uncooked, or improperly washed vegetables and fruits. The contact of Vegetables and fruits with dust, soil and waste water have played a major role as a contamination source since soil and waste water harbors millions of parasites and bacteria. So, Consideration has to be given to the bad habit of the Ethiopian custom of consuming raw fruits and vegetables without treating with insufficient heat or detergents as nationwide. Because vegetables and fruits which are sold in the market can bring other side health risks since it mostly acts as a reservoir for many microorganisms.

2.3 Knowledge attitude and practice towards food safety

Previous studies from different regions of the world showed that knowledge towards food safety is low(44,45). Food safety attitude is a crucial aspect that may influence food safety performance and practice, accordingly lessening the happening of foodborne illnesses(46). A previous study from Ethiopia found that, only 29.1% of the study participants had a positive attitude toward food safety. This study also found that there were significant differences in attitudes towards food safety due to their variation in the level of knowledge (47).This finding was supported by other studies, which revealed that there was a significant association between knowledge level and attitude score. However, this result was contradicted by another study. On the other hand, there was no substantial correlation between food safety attitude and food safety practice. This finding was in line with another study conducted in Malaysia (48). Other studies have shown that positive attitudes motivate food handlers to have a greater effect on their food safety practices (49), and it is more significant than knowledge and practice (15).

2.3. Conceptual Frame work

The proposed conceptual framework builds on existing evidence that addresses factors that affect parasitic and bacterial contamination of fruits and Vegetables sold in open aired markets. It shows how dependent and independent variables are related each other.

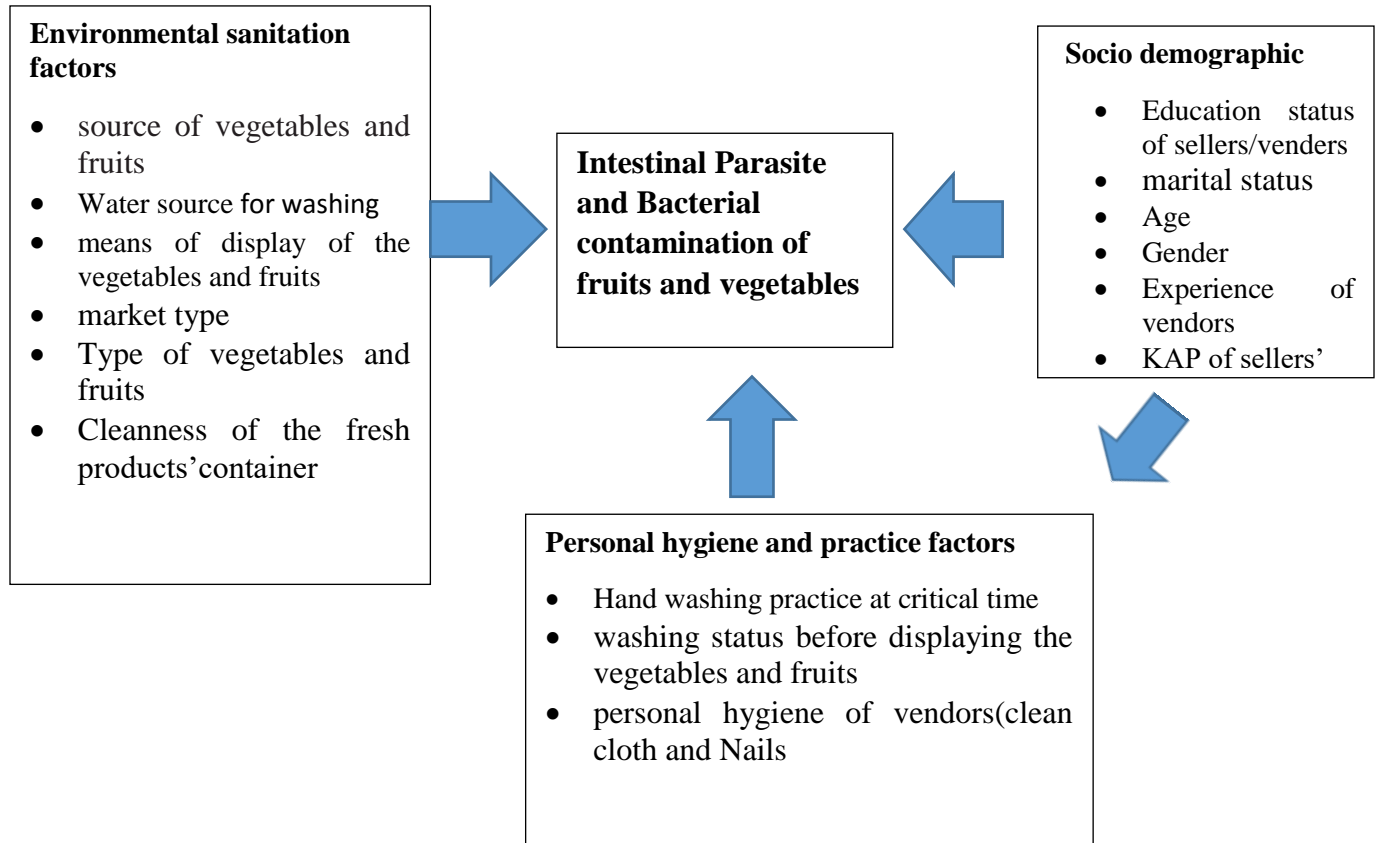


Figure 1: Conceptual framework of study on Intestinal Parasitic and Bacterial contamination of Fruits and Vegetables sold in open aired markets at peri-urban Kebeles of Jimma town Southwest Ethiopia 2021.

CHAPTER THREE

3. OBJECTIVE OF THE STUDY

3.1 General Objective

- ❖ To determine prevalence of intestinal parasitic and bacterial contamination of fruits and vegetables sold in open aired markets and assess KAP of sellers in peri-urban Kebeles of Jimma town, Southwest Ethiopia, 2021.

3.2 Specific objectives

- ❖ To determine prevalence of intestinal parasites contamination of fruits and vegetables sold in open aired markets in peri-urban Kebeles of Jimma town, 2021.
- ❖ To determine prevalence of bacterial contamination of fruits and vegetables sold in open aired markets in peri-urban Kebeles of Jimma town, 2021.
- ❖ To assess antibiotic resistance of bacterial species isolated from fruits and vegetables.
- ❖ To assess factors associated with intestinal parasitic and bacterial contamination of fruits and vegetables sold in open aired markets in peri-urban Kebeles of Jimma town, 2021.
- ❖ To assess KAP (knowledge, attitude and Practice) of sellers about intestinal parasitic and bacterial contamination of fruits and vegetables sold in open aired markets in peri-urban Kebeles of Jimma town, 2021.

CHAPTER FOUR

4. METHERIALS ANDMETHODS

4.1. Study Area and Study Period

The study was conducted in three open aired markets (Hora Gibe/Tishayer, Bore and Jiren markets) from Hora Gibe Kebele, Bore, and Jiren kebeles which are peri-urban Kebeles of Jimma Town Southwest Ethiopia from July 1-September 1, 2021. Jimma is the largest town in Southwestern Oromia region in Ethiopia. It is located in Southwestern part of Ethiopia and about 352 km distant from Addis Ababa, the capital city of Ethiopia. The town is located at 7° 40' North latitude and 36° 5' East Longitude and the climate condition is relatively cool tropical monsoon climate, average altitude of about 1780 m above sea level, a mean annual maximum temperature of 30°C and a mean annual minimum temperature of 14°C and annual rainfall ranges from 1138 mm to 1690 mm.

Projected Population for the year 2020/2021 showed that a total of 229,157 inhabitants live in Jimma town (50) According to Jimma town administration health office there are about 23 governmental health care institutions in Jimma town. Among them one referral hospital, one general hospital, four health centers and 17 health posts exist. The total health care coverage in the city was 50% at hospital level, 76% at health center.

Jiren Kebele is one of study area that has 7,138 total populations from these 3,589 are males and 3,549 are females and the kebele has 1,487 household sizes and it has one market. Bore Kebele is the other study area that has 5,970 total populations from these 3,002 are males and 2968 are females and the kebele has 1,244 households' size and it has one market. Hora Gibe Kebele has 1,651 total populations from these 830 are males and 821 are females and the kebele has 344 households' size and it has one market. Kofe kebele has 2,855 total populations from these 1,435 are males and 1,419 are females and the kebele has 595 households' size and it hasn't market. Efa Bula kebele has 7,731 total populations from these 3,887 are males and 3844 are females and the kebele has 1,611 households size and it hasn't market.

4.2 Study Design

A community based cross--sectional study design was conducted.

4.3 Source Population

The source population for this study were all sellers who sales fruits and vegetables in open aired markets and all fruits and vegetables that were sold in open aired markets of Peri-urban Kebeles of Jimma town from July 1 to September1, 2021.

4.4 Study Population

The study populations were selected sellers, and fruits and vegetables those sold in peri urban kebele market places i.e Jiren, Bore and Hora Gibe Kebeles from July 1 to September1, 2021.

4.5 Study Unit

The study unit was selected sellers who sale fruits and vegetables in open aired markets and selected fruits and vegetables that were sold in open aired markets from July 1 to September1, 2021.

4.6 Inclusion and Exclusion Criteria

Inclusion Criteria:All sellers who sales fruits and vegetables in open aired markets in peri urban kebeles of Jimma town during the study period and all fruits and vegetables those sold and bought in open aired markets during data collection were included in the study.

Exclusion Criteria: Sellers who sale fruits and vegetables in open aired markets in peri urban kebeles of Jimma town during the study period but, unwilling to answer for question of questioner due to different problems or reason were excluded from the study.

4.7. Sample size Determination for fruits and vegetables

The sample size required for the study calculated using the formula to estimate a single population proportion by considering the following assumptions:

$$N = \frac{(Z\alpha/2)^2 P(1-P)}{d^2}$$

Where,

- ✓ **Assumptions:** With the assumptions of 95% Confidence level,
- ✓ **n=** required sample size,
- ✓ $Z\alpha/2$ = critical value for normal distribution at 95% confidence level which equals to 1.96 (Z value at alpha=0.05),
- ✓ **P=** Established prevalence of IP contamination of fruits and vegetables
- ✓ **d=** desired precision (5% margin of error).

Based on the previous study the overall contamination rate was (P=57.8%) (28, 10),the sample size (N) was calculated as

$$N = \frac{(1.96)^2 * 0.578(1-0.578)}{0.05^2} = 375$$

So, total sample size was 375 fruits and vegetables and 375 sellers.

4.9. Sample Collection Technique for both Parasites and Bacterial tests

Study subjects were selected by simple random sampling for KAP of sellers'. Eight types of vegetables and fruits frequently consumed by peri-urban kebeles communities (lettuce (46), cabbage (47), carrot (47), tomato (47), green pepper (47), banana (47), mango (47) and avocado (47)) were collected from local markets in Jiren, Bore and Hora Gibe Kebeles of Jimma town. Each vegetable or fruit type was purchased under normal condition from randomly selected sellers from each market (Figure 1).

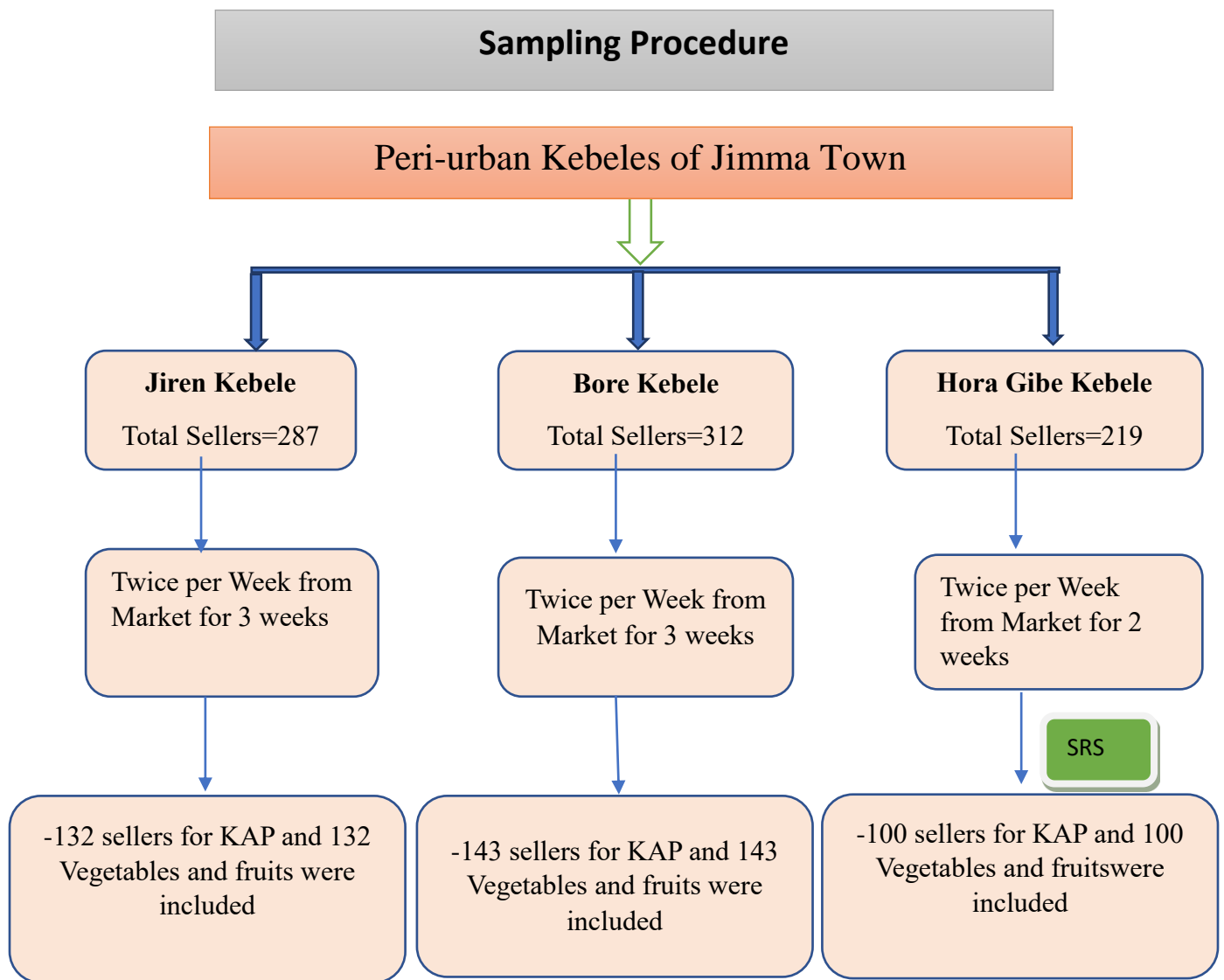


Figure 2: A diagram indicating sampling procedure

Each sample was collected into a sterile polythene bag and labeled with information such as sample code, sample type, market name and date of collection. Moreover, information such as market type, source of the vegetables and fruits, whether the vegetables and fruits were washed before display, the educational status of the sellers, marital status of the sellers and experience of the sellers was recorded for each sample on questionnaire. The collected samples were transported immediately to Jimma University Medical Parasitology and microbiology Laboratory for further processing.

4.8.1. Sample processing for isolation of Parasites

About 300g of fruits and vegetables were bought and a portion (200 g) of each vegetable and fruit was washed separately in 500 mL of normal saline for detaching the parasitic stages (ova, larvae, cysts, and oocysts) of helminths and protozoan parasites commonly assumed to be associated with vegetable contamination. After overnight sedimentation of the washing solution, 15 mL of the sediment was transferred to a centrifuge tube using sieve, to remove undesirable matters. For concentrating the parasitic stages, the tube was centrifuged at 3000 rpm for five minutes(26). After centrifugation, the supernatant was decanted carefully without shaking. Then the sediment was agitated gently by hand for redistributing the parasitic stages. Finally, the sediment was examined under a light microscope using $\times 10$ and $\times 40$ objectives. Modified Zeihl-Neelsen staining technique was used for identification of oocysts of *Cryptosporidium*, *Cystoisospora*, and *Cyclospora spp* as described elsewhere (51). [ANNEX-IV]

4.8.2. Sample processing for Isolation of bacteria

About 300g of fruits and vegetables samples were transported in sterile polythene bag and analyzed for isolation and identification of pathogenic bacteria following standard methods. About 25g of vegetable and fruit samples were rinsed thoroughly with 225 ml sterile distilled water/sterile 0.1%(W/V) bacteriological peptone water (oxid) for 3 minutes and tenfold serial dilutions of 10^{-1} , 10^{-2} , 10^{-3} , 10^{-4} and 10^{-5} was made and then 0.1 ml of the suspension from each dilution was pipetted into Mac Conkey agar, Salmonella - Shigela agar, and Mannitol Salt agar using the streak Plate Technique. The plates were allowed to solidify, invert and incubate at 37 °C for 24 h for colony formation. Distinctive morphological properties of each pure culture such as colony form, elevation of colony and colony margin were observed. Bacterial specieses were isolated and identified by gram stain, cultural and different biochemical tests following standard protocol as described elsewhere (47)[ANNEX-IV].

4.8.3. Antibiotic Susceptibility Testing of Bacteria Isolates

Pathogenic Bacterial that isolated suspension was inoculated on Muller Hinton agar using commercial discs (Himedia, India) medium using sterile swab stick. Commercially prepared, fixed-concentration filter-paper antibiotic discs were applied on the plate surface. Disc diffusion technique was used to determine the drugs sensitivity of the isolated bacteria.

The plates were inverted and incubated aerobically at 37⁰C for 24 hours. The antibiotics used were Ciprofloxacin (5µg), Gentamycin (10µg), Ceftriaxone(30µg), Piperacillin-tazobactam(100/10µg), Erythromycin(E-15µg), Trimethoprim-sulfamethoxazole(co-trimoxazole) (1.25/23.75µg), Meropenem (10µg), Ampicillin (AMP- 10µg), and cefazolin (30 µg), Tobramycin(10µg), Cefoxitin(30µg), Penicillin(10µg), Clindamycin(2µg), Amoxicillin-clavulanate(20/10µg). Standard reference strains *E. coli* (ATCC 25922), *S. aureus* (ATCC 25923) and *Salmonella spp*(ATCC13311) were used for quality control of the antibiotics. Sensitivity of the isolated bacteria was determined according to the Clinical and Laboratory Standards Institute guideline (52).

4.9. Study variables

4.9.1. Dependent variables

Prevalence of medical important intestinal parasites and bacteria contamination of fruits and vegetables and KAP of sellers

4.9.2. Independent variables

Type of vegetables and fruits, source of vegetables and fruits, means of display of the vegetables and fruits, washing status before displaying the vegetables and fruits, water source for washing, personal hygiene of vendors (clean cloth and Nails), Cleanness of the fresh products' container and educational status, marital status, experience, age, gender of the sellers or vendors.

4.10 Operational Definitions

Intestinal parasite positive status: if the study samples have one of the parasites' eggs, cyst, trophozoite, larva or more than one of this diagnostic stage of intestinal parasites was found in the samples.

Parasitic contamination: if the fruits and vegetables contaminated by Intestinal parasites.

Bacterial contamination: if the fruits and vegetables contaminated by bacteria.

Open-aired markets: a public marketplace where foods and goods is sold and bought.

Fresh vegetables andfruits: vegetables that have not been processed in any manner

Raw vegetables: Vegetables that uncooked

Adequate personal hygiene: personal hygiene of vendors includes the use of clean cloth and the finger nails are clean, and short

Inadequate personal hygiene: personal hygiene of vendors includes the use of unclean cloth and the finger nails are not clean.

Attitude: The way asellers thinks and behaves to ward intestinal parasitic and bacterial contamination of fruits and vegetables, itwas measured by 9 questions with five point Likert's scale in this study.

Positive Attitude: Sellers who were responded above the mean value for attitude questions (if Participantswerescore > mean score).

Negative Attitude: Sellers who were responded below the mean value for attitude questions (if Participantswerescore <mean score).

Knowledge: It is the awareness of the sellers about intestinal parasitic and bacterial contamination of fruits and vegetables; it was measured by 13 questions.

Good knowledge/knowledgeable: Sellers who were responded above the mean value for knowledge questions (if Participants were score > mean score of the correctly answer questions).

Poor knowledge/not knowledgeable: Sellers who were responded below the mean value for knowledge questions (if Participants were score <mean score of the correctly answer questions).

Practice: Assessment of seller's actual activities/habitual involvement to prevent intestinal parasitic and bacterial contamination of fruits and vegetables, it was measured by 9 questions.

Good Practice: Sellers who were responded/scored above the mean value for practice questions (if Participants were score > mean score of the correctly answer questions).

Poor/bad Practice: Sellers who were responded/scored below the mean value for practice questions (if Participants were score <mean score of the correctly answer questions).

4.11 Data Collection Tool

Questionnaire

Data was collected using a semi-structured questionnaire based interview, which is adopted from (53,54) and previously done studies and modified in the form that will answer the study objectives. The questionnaire was prepared in English language. Samples (fruits and Vegetables) were bought from vendors and variables data was collected by interview by using structured questionnaire for assessment of KAP of vendors about contamination and microscopic laboratory examination for intestinal parasitic and Culture and biochemical test for Bacteria isolation. The questionnaire contains four parts; the first part assesses the sociodemographic characteristics of the vendors; the second part of the questionnaire was to assess the knowledge of the vendors towards the safety of fruits and vegetables. This part comprises of 13 multiple-choice questions with 'True', 'false', 'Don't know options/ don't remember' options. The third part of the questionnaire contains 9 questions to assess vendors' attitude towards fruits and vegetables safety with scale rating 'Extremely Disagree, disagree, neutral, agree and extremely agree'. The fourth part of the questionnaire includes 9 questions to assess vendors' practice of fruits and vegetables safety with response options of 'Never, rarely, sometimes, most of the times and always.'

4.12. Data Quality Assurance

The quality assurance of the study was started at the very beginning of study instrument development. Data collection instrument was developed based on the previously studies and discussed variables. Question of questionnaire was translated to Afan Oromo and Amharic language and back to English for its consistency. Before actual data collection, the data collectors were trained and had a similar concept on the questionnaire. The questionnaire was

tested by pretest to identify errors and modify it. During data collection, close supervision was performed. Based on the challenges and errors detected during data collection, frequent meeting was held to overcome it. AST disc, Reagent, Culture Media were checked. The collected data was checked for completeness and correctness of the information before analysis. Data was coded and missed data was managed. All the laboratory investigation was performed based on standard operating procedures (SOP). Samples were processed immediately after bought from markets to decrease the errors. Before examination of the samples, internal quality control was done to assure the materials, reagents, culture media and antibiotic susceptibility testing disk were well to process examination. Data entry and analysis soft ware was checked with first few collected data to assess the correctness of the prepared template and weather it processes what was wanted.

4.12.1 Pre-test study

A pre-test study was conducted by semi-structured questionnaire to some nearby kebele sellers before starting data collection. The pre-test was to test the clarity of the items in the questionnaire tools, the time needed to answer the questions and to identify any difficulties that may arise and need to be clarified before applying the questionnaire for actual data collection.

4.13 Data Processing and Analysis

Data entry was performed by using Epi-data 4.6 version software and for data analysis transferred to SPSS 25.0 statistical software. Different frequency tables, graphs and descriptive summaries were used to describe the study variables. Logistic regression analysis was used to see significance of association between dependent and independent variables. Variables with a p-value of 0.25 or less were further analyzed by multivariate analysis to identify significant factors for dependent variable. Variables with P-value < 0.05 were considered as statically significant

4.14 Ethical Consideration

Prior to the study implementation, ethical clearance was obtained from Institutional Ethics Board (IRB) of the JU Institute of Health, Faculty of Health Sciences. Supportive letter was obtained from School of Medical Laboratory Science, Department of Medical Parasitology. Consultation and permission to conduct this study was also obtained from Jimma town health office as appropriate. Copy of the Ethical clearance letter was given to Jiren, Bore and Hora Gibe Kebele Administrative which was asked for its cooperation, and then the official

letter was gotten from the three Kebeles to collect data. Prior to be started the study; verbal consent was obtained from the Vendors/participants.

Throughout the study, the data was confidentially protected and anonymity of the study participants. Interview was conducted in the separated area, enclosed whenever possible to protect the study participants' privacy. In order to protect the study participants' identities, each participant was given the unique identification code, which was checked before transcription of the data.

4.15 Dissemination and Utilization of Results

The findings of this study will be presented to Jimma University, Faculty of Health Sciences, distributed to Jimma town health department, for concerning bodies and to other organizations working on related area. The findings may also be presented in different seminars, meetings and workshops. It will be published in national/international journal.

CHAPTER FIVE

5. RESULTS

4.1 Socio-demographic and Environmental characteristics

A total of 375 Sellers/vendors comprising of 342 females and 33 males were participated in the current study of intestinal parasitic and bacterial contamination of fruits and vegetables. Sellers mean age was 29.02 ± 4.99 years while their average monthly income was 1963.20 ± 391.87 ETB. One hundred forty-three (38.1%) of sellers were from Bore kebele Bore market, 132(35.3%) were from Jiren market and the rest 100(26.7%) were from Hora Gibe Kebele: Tishayer market. The majority of the Sellers were attended primary education 181(48.3%), and 130(34.7.5%) had secondary education. Regarding sellers experience almost half of them had 3-5 years' experience. One hundred forty-three of fruits (N=72) and vegetables (N=71) were collected from Bore, 100(26.7%) fruits (N=50) and vegetables (N=50) from Hora Gibe and the rest 132(35.2%) fruits (N=66) and vegetables (N=66) were from Jiren. Regarding the source where sellers brought vegetables and fruits, 212(56.5%), bought from farmers and 10(2.7%) from their private garden but in 150(40.8%) of cases there were middle men. One hundred eighteen (31.5%) of vendors had adequate personal hygienic condition while 257(68.5%) of them didn't have adequate hygienic condition during data collection time. Sellers were asked weather they wash or not the vegetables/fruits before display and 215(57.3%) of them replied no and the rest 160(42.7%) of them replied as they would do. Majority (288. 76.8%) of sellers display fruit and vegetables in the floor while the rest 87(24.2%) of them display on the table/shelf for sale (Table 1).

Table 1: Socio-demographic and characteristics of the study participants in peri urban kebeles of Jimma town, Jimma zone, from July-September 2021.

Variables	Category	Frequency	Percentage
Gender of sellers	Male	33	8.8
	Female	342	91.2
Age of sellers	<19 years	5	1.3
	20-29 years	222	59.2
	30-39 years	134	35.7
	40 and above years	14	3.7
Educational status	No formal education	58	15.5
	Primary education	181	48.3
	Secondary education	130	34.7
	Higher education	6	1.6
Marital status	Single	83	22.1
	Married	262	69.9
	Divorced	19	5.1
	Widowed	11	2.9
Average monthly income	<1500ETB	60	16.0
	1500-2000ETB	177	47.2
	2000-2500ETB	123	32.8
	2500-3000ETB	14	3.7
	>3000ETB	1	0.3
Experience of sellers	<1	21	5.6
	1-2	89	23.7
	3-5	184	49.1
	>5	81	21.6
Market sites of sample collection	Bore	143	38.1
	Hora Gibie	100	26.7
	Jiren	132	35.2
Source of vegetable	Farmers	212	56.5
	Middle Men	153	40.8
	Private Garden	10	2.7
Sellers' hygiene condition	Adequate	118	31.5
	Inadequate	257	68.5
Cleanness of the containers	Adequate	101	26.9
	Inadequate	274	73.1
Wash status	Yes	160	42.7
	No	215	57.3
Source of water	Pipe	24	3.75
	Well	99	61.9
	River	37	3.74
Means of display	floor	288	76.8
	Shelf	57	15.2
	table	30	8.0

ETB refers to: Ethiopian birr and SD: standard deviation

5.2 Parasitic Contamination

5.2.1 Prevalence of Parasitic Contamination

Of the total of 375 samples analyzed, 173 samples were found contaminated with at least one type of parasite making prevalence of intestinal parasites 46.1%. From this only in 1.1 % of the samples more than one type of intestinal parasite species were isolated. From the total 375 samples 187 (49.9%) of the samples were vegetables and the rest 188(50.1%) were fruits. From 187 vegetables examined, 105(60.6%) were found to be contaminated at least one type of parasite while from 188 fruits, 68 (39.4%) were contaminated. The parasite detected includes ova of *A. lumbricoides*, *Toxocara spp.*, *Hymenolepis nana*, *Fasciola spp* and oocysts *Cyryptosporidium spp*; and cysts of *G. intestinalis*, *E. histolytica/dispar* and, *Strongyloides stercoralis*. *A.lumbricoides* 46(12.3%) was the most frequently detected parasite, followed by *Strongyloides stercoralis* 32(8.5%), *E.histolytica/dispar* cyst 30(8%), *Hymenolepis nana* 23(6.1%), *G. intestinalis* cyst 22(5.9%), oocysts *Cryptosporidium spp* 12(3.2%), *Toxocara spp* 10(2.7%), and *Fasciola spp* 2(0.5%) (Table 3).

Table 2: Frequency of Distribution of Parasitic Contaminations Among Fruits and Vegetables Sold in open aired markets in peri urban kebeles of Jimma Town from July-September 2021. (N=375)

Kind of Item Product	Number Examined	Number Positive (%)	Parasite Species Detected, N (%)		
			One	Two	
Vegetables	Lettuce	46(12.3)	29(63.1)	28(60.9)	1(2.2)
	Cabbage	47(12.5)	29(61.7)	29(61.7)	0
	Carrot	47(12.5)	30(63.8)	30(63.8)	0
	Green pepper	47(12.5)	17(36.1)	16(34)	1(2.1)
	Total	187(49.9)	105(60.6)	103(59.4)	2(1.2)
Fruits	Tomato	47(12.5)	23(48.9)	22(46.8)	1(2.1)
	Banana	47(12.5)	7(14.9)	6(12.8)	1(2.1)
	Mango	47(12.5)	18(38.3)	18(38.3)	0
	Avocado	47(12.5)	20(42.6)	20(42.6)	0
	Total	188(50.1)	68(39.4)	66(38.2)	2(1.2)
Overall distribution	375	173(46.1)	169(45.1)	4(1.1)	

The highest prevalence of parasites was detected in carrot 30 (63.8%), succeeded by lettuce 29(63.1%) and Cabbage 29(61.7%). Banana with 7(14.9%) prevalence was the least contaminated fruit. *Strongyloides stercoralis* was the most frequent parasite detected in carrot. Out of 343 fruits and vegetables samples purchased from open market 171 were positive for at least one type of parasite and from 32 sample purchased from Grocery 2 were positive for at least one type of parasite. The most frequently encountered parasitic egg/cyst was *A. lumbricoides* 46(12.3%), followed by *Strongyloides stercoralis* 32(8.5), *E. histolytica/dispar* cysts 30(8%) and the least occurrence was *Fasciola* spp 2(0.5%) (Table 3).

Table 3: Distribution of Intestinal parasites contamination in fresh fruits and vegetables collected from open aired markets in Peri Urban Kebele of Jimma Town from July-September, 2021

DETECTED PARASITES	VEGETABLES	FRUITS	TOTAL (N=375)
	N (%)	N (%)	N (%)
<i>A.lumbricodes</i>	30(12.8))	16(4.3)	46(12.3)
Stroglyoides Like Larva	23(9.8))	9(2.4)	32(8.5)
<i>H. nana</i>	16(6.8))	7((1.9)	23(6.1)
<i>E.histolytica/dispar cyst</i>	25((10.6)	5(1.3)	30(8)
<i>G.lambliia cyst</i>	17(7.2)	5(1.3)	22(5.9)
<i>Cyryptosporidium Spp</i>	9(3.8)	3(0.8)	12(3.2)
<i>Toxocara Spp</i>	9(3.8)	1(0.3)	10(2.7)
<i>Fasciola Spp</i>	2(0.08)	0	2(0.5)

Among 173 samples which were found positive for intestinal parasites, the highest prevalence of parasite was found in samples collected from Bore (66, 38.15%) followed by Jiren (60, 34.7%) and least was found from samples collected from Hora gibe (47, 27.16%)

As it is shown in the following figure (*figure 3*) there is equal distribution of parasites among fruits and vegetables collected from Bore while the prevalence is higher in vegetables collected from Jiren and Hora gibe compared to fruits collected from these areas.

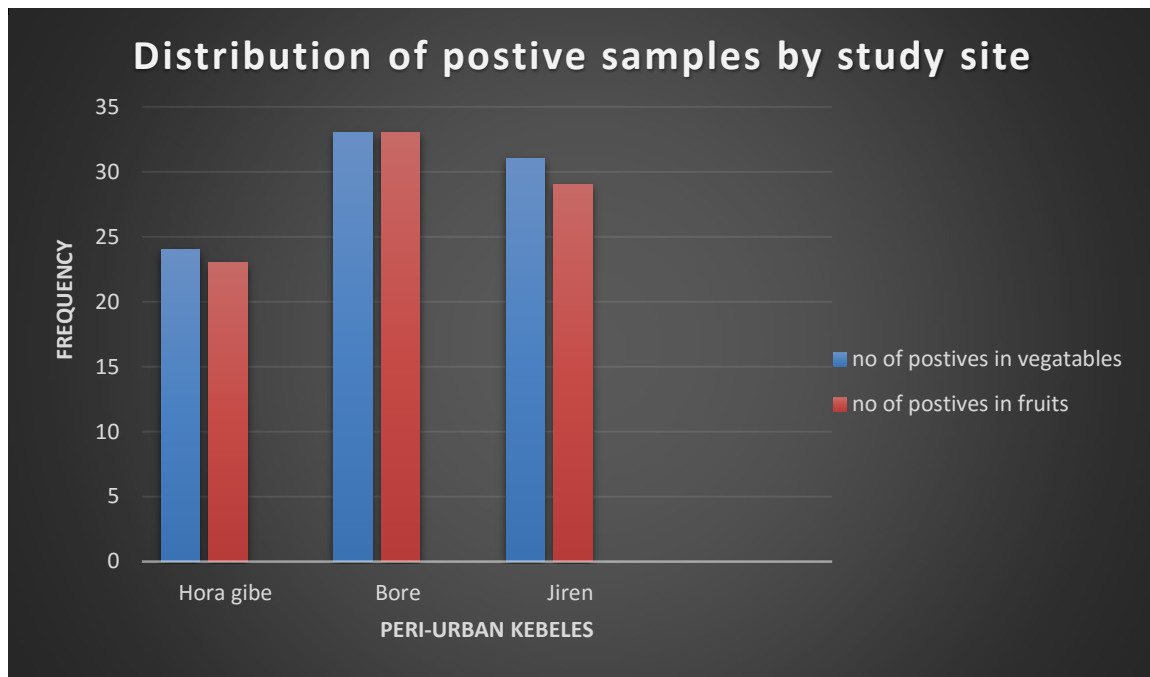


Figure 3: Distribution of positives in fruits and vegetables among study sites

5.2.2. Factors affecting parasitic contamination

To identify associated factors parasitic contamination bivariate and multivariate logistic regression analysis were performed.

Accordingly, bivariate analysis was performed to avoid confounding effect before performing multivariate analysis and variables whose p-value was < 0.25 were considered as candidate variable for multivariate analysis. Among fifteen variables were entered in SPSS for binary logistic regression and eleven variables were candidate for multiple logistic regression (P-value < 0.25)

Table 4: Bivariate analysis using logistic regression analysis output showing factors affecting parasitic contamination of fruits and vegetables

Variable	Category	Parasitic contamination		COR(95% CI)	P-value
		Yes	No		
		Freq (%)	Freq (%)		
Sex of sellers	Female	179(47.7)	163(43.5)	0.23(0.09-0.58)	0.002*
	Male	6(1.6)	27(7.2)	1	
Educational status of sellers	No formal education	53(14.1)	5(1.3)	52.08(18.77-144.52)	<0.001*
	Primary	97(25.9)	84(22.4)	5.67(1.07-3.32)	<0.001*
	Secondary and above	23(6.1)	113(30.1)	1	
Age of sellers	<19	2(0.5)	3(0.8)	0.7(0.08-5.30)	.702
	20-29	79(21.1)	143(28.1)	0.6(0.19-1.63)	.283
	30-39	85(22.7)	49(13.1)	1.74(0.58-5.24)	.329
	40 and above	7(1.9)	7(1.9)	1	
Experience of sellers	<1 year	15(4)	6(1.6)	13.08(4.28-39.97)	<0.001*
	1-2 year	62(16.5)	27(7.2)	12.01(5.7-25.32)	<0.001*
	3-5	83(22.1)	101(26.9)	4.3(2.22-8.32)	<0.001*
	>5	13(3.5)	68(18.1)	1	
Marital status of sellers	Single	26(6.3)	50(13.9)	1	
	Married	124(33.3)	130(34.7)	1.83(1.07-3.13)	0.026*
	Divorced	13(3.5)	13(3.5)	1.92(0.78-4.75)	.156*
	Widowed	10(2.7)	9(2.4)	2.18(0.77-5.9)	.144*
Site collection	Bore	66(17.6)	77(20.5)	1.03(0.6-1.65)	0.90
	Hora Gibe	47(12.5)	53(14.1)	1.06(0.63-1.79)	0.82
	Jiren	60(16.0)	72(19.2)	1	
Type of market	Grocery	2(0.5)	30(8.0)	1	

	Open market	171(45.6)	172(45.9)	14.91(3.51-63.38)	<0.001*
Source of vegetables and fruits	Farmers	113(30.1)	99(26.4)	1.14(0.32-4.06)	0.838
	Middle men	55(14.7)	98(26.1)	0.56(0.561-2.024)	0.377
	Private garden	5(1.3)	5(1.3)	1	
Hygienic condition of sellers	Adequate	19(5.1)	99(26.5)	1	
	Not adequate	154(41.1)	103(27.5)	7.79(4.49-13.51)	<0.001*
Wash status	Yes	8(2.1)	152(40.5)	1	
	No	165(44.0)	50(13.3)	62.7(28.79-135.53)	<0.001*
Type of Products	Vegetables	105(28)	82(21.9)	2.99(1.94-4.61)	<0.001*
	Fruits	68(18.1)	120(32)	1	
Means display	Floor	167(44.5)	121(32.1)	18.63(7.872-44.1)	<0.001*
	Table/Shelf	6(1.6)	81(21.6)	1	
Knowledge	Good Knowledge	111(29.6)	54(14.4)	1	
	Poor Knowledge	62(16.5)	148(39.5)	4.91(3.16-7.62)	<0.001*
Attitude	Positive attitude	57(15.2)	138(36.8)	1	
	Negative attitude	116(30.9)	64(17.1)	4.38(2.84-6.77)	<0.001*
Practice	Good practice	17(4.5)	135(36.0)	1	
	Poor practice	156(41.6)	67(17.9)	18.49(10.35-33.02)	<0.001*

Finally, multiple logistic regression analysis was performed by putting all the eleven variables which were candidate by bivariate analysis using SPSS by enter method and variables whose P-value is less than 0.05 were fitted in the final model. Among eleven variables entered in multiple logistic regressions only six variables (fruits and vegetables washed before display, Type of Products, means of display, experience, educational status and practice of sellers) showed statistically significant association with parasitic contamination.

By putting all other variables constant the likelihood of parasitic contamination of fruits and vegetables is 251.775 times in fruits and vegetables not washed than washed before display (95% C.I. 53.387-1187.387, P-value= <0.001), 8.516 times in vendors/ sellers who had 1-2 years of experience than who had more than five years (C.I, 1.146-63.30, P-value= 0.036), 5.782 times in vegetables than fruits (C.I. 1.658-20.170, P-value = 0.006), 54.766 times in fruits and vegetables displayed on the floor than on the table/shelf (C.I. 10.108-296.733, P-value= <0.001), 21.734 times in fruits and vegetables collected from vendors who had no formal education than who had secondary and above education (C.I 1.677-281.609, P-value =0.018) and 6.343 times in fruits and vegetables collected from vendors who had poor practice regarding handling of fruits and vegetables than those who had good practice (C.I. 1.767-22.770 P-value = 0.005). In table 16 below multiple logistic regressions output is presented including AOR with 95% C. I and P-value of all variables entered.

Table 5: Multiple logistic regression out put

Variable	Category	AOR (95% CI for AOR)	P-value
Sex of sellers	Female	4.325(0.418-44.796)	.219
	Male	1	
Market type	Grocery	1	
	Open Market	0.359(0.023-5.651)	.466
Wash status	Yes	1	
	No	251.775(53.387-1187.387)	<0.001*
Marital status of sellers	Single	1	
	Married	4.326(0.909-20.590)	.066
	Divorced	0.353(0.032-3.840)	.393
	Widowed	6.222(0.321-120.658)	.227
Experience of sellers	<1 year	9.924(0.635-155.068)	.102
	1-2 year	8.516(1.146-63.302)	.036*
	3-5 years	1.594(0.314-8.093)	.574
	>5 years	1	
Hygienic condition	Adequate	1	
	Not adequate	2.512(0.565-11.163)	.226
Type of product	Vegetables	5.782(1.658-20.170)	.006*
	Fruit	1	
Educational status	No formal education	21.734(1.677-281.609)	.018*
	Primary education	1.478(0.384-5.684)	.570
	Secondary and above	1	
Means of display	On the floor	54.766(10.108-296.733)	<0.001*
	On the table/shelf	1	
Attitude	Negative	1.113(0.327-3.797)	.864
	Positive	1	

Knowledge	Good	1	
	Poor	0.327(0.087-1.225)	.097
Practice	Good	1	
	Poor	6.343(1.767-22.770)	.005*

AOR refers to Adjusted Odds ratio, 1 refers to reference group and * refers to variables showed statistically significant association with dependent variable.

5.3 Bacterial contamination

5.3.1 Prevalence of bacterial contamination

In the present study, besides intestinal parasite contamination, bacterial contamination of Fruits and Vegetables were examined. As a result, from the total of 375 fruit and vegetable samples collected from open aired markets in 194 (51.7%) fruit and vegetable samples were contaminated with at least one bacterium. Nine species of bacteria were detected *S. aureus* being the commonest bacterial contaminant of fruits and vegetables (77, 20.5%) followed by salmonella species (66, 17.6%). In 194 (51.7%) fruit and vegetable samples at least one bacterial species was isolated while 44(11.7%) of samples were contaminated with two bacterial species. Only in 3(0.8%) of samples three bacterial species were found. (Table 6).

Table 6: Frequency of Distribution of Bacteria Contaminations Among Fruits and Vegetables Sold in open aired markets in peri urban kebeles of Jimma Town from July-September 2021. (N=375)

Type of vegetable/fruit	Number examined	Number contaminated	Single and Multiple bacteria Species Detected, N (%)		
			Single	Double	Triple
Lettuce	46	26(56.52)	20(43.47)	6(13.04)	0
Cabbage	47	22(46.8)	17(36.17)	4(8.5)	1(2.1)
Carrot	47	27(57.44)	21(44.68)	6(12.76)	0
Tomato	47	15(31.9)	13(27.65)	1(2.1)	1(2.1)
green pepper	47	23(48.93)	17(36.17)	6(12.76)	0
Avocado	47	32(68.08)	22(44.68)	11(23.4)	0
Banana	47	22(46.8)	17(36.17)	5(10.63)	0
Mango	47	27(57.44)	21(44.68)	5(10.63)	1(2.1)
Total	375	194(51.7)	147(39.2)	44(11.73)	3(0.8)

Regarding distribution of Bacteria species among fruit and vegetable type, Avocado was the most frequently contaminated fruits by single and double bacteria species (44.93.6%) followed by Mango (34, 72.3%) and Carrot (33, 70.2%), and Tomato with (18, 38.29%) prevalence was the least contaminated fruit. Generally, contamination was higher among fruits compared to vegetables 30.1%) [Table 7]

Table 7: **Distribution of bacterial contamination among fruits and vegetables sold in open aired markets of peri-urban kebeles of Jimma Town from July to September 2021.**

vegetable/fruit	Category	Number examined	Number contaminated with each species of bacteria					
			E. coli N (%)	Salmonella spp. N (%)	Shigella spp. N (%)	Klebsiella spp. N (%)	S. aureus N (%)	Others N (%)
Vegetable	Lettuce	46	5(11)	12(26)	3(6.5)	4(8.7)	8(17.4)	0
	Cabbage	47	7(14.9)	6(12.8)	1(2.1)	3(6.4)	9(19.1)	2(4.3)
	Carrot	47	8(17.0)	10(21.3)	1(2.1)	5(10.6)	9(19.1)	0
	green pepper	47	3(6.4)	7(14.9)	3(6.4)	9(19.1)	8(17.0)	0
	Total	187	23(12.3)	35(18.7)	8(4.3)	21(11.2)	34(18.2)	2(1.1)
Fruit	Tomato	47	6(12.8)	6(12.8)	0	3(6.4)	2(4.3)	1(2.1)
	Banana	47	3(6.4)	8(17.0)	1(2.1)	2(4.3)	13(27.7)	0
	Mango	47	5(10.6)	6(12.8)	3(6.4)	5(10.6)	12(25.5)	3(6.4)
	Avocado	47	6(12.8)	11(23.4)	3(6.4)	7(14.9)	16(34.0)	1(2.1)
	Total	188	20(10.6)	31(16.5)	7(3.7)	17(9.0)	43(22.9)	5(2.6)
Total		375	43(11.5)	66(17.6)	15(4.0)	38(10.1)	77(20.5)	11(2.9)

Others refers to: *Providencia spp*, *Enterobacter spp*, *Proteus spp* and *Citrobacter Spp*

5.3.2 Associated factors with bacterial contamination

To identify factors affecting contamination of fruits and vegetables by bacteria bivariate and multivariate logistic regression analysis were performed. Accordingly, bivariate analysis was performed to avoid confounding effect before performing multivariate analysis and variables whose p-value was < 0.25 were considered as candidate variable for multivariate analysis. Among seventeen variables entered in SPSS for binary logistic regression thirteen variables were candidate for multiple logistic regression (P-value <0.25). Table 8 below shows bivariate analysis output by binary logistic regression.

Table 8 : Bivariate analysis showing factors associated with bacterial contamination of fruits and vegetables

Variable	Category	Bacterial contamination		COR	95% CI for COR		P value
		Yes	No		Lower	Upper	
		Freq (%)	Freq (%)				
Sex of sellers	Female	179(47.7)	163(43.7)	0.759	.370	1.555	0.451
	Male	15(4.0)	18(4.7)				
Educational status of sellers	No formal education	34(9.1)	14(6.4)	2.024	1.084	3.778	<0.001*
	Primary	104(27.7)	77(20.5)	1.929	1.229	3.029	<0.001
	Secondary and above	56(14.9)	80(21.3)				1
Age of sellers	<19 years	3(0.8)	2(0.5)	1.500	.189	11.927	0.702
	20-29 years	111(29.6)	111(29.6)	1.000	.340	2.945	0.99
	30-39 years	73(19.5)	61(16.3)	1.197	.398	3.600	.749
	40 and above years	7(1)	7(1)				1
Experience of sellers	<1 year	12(3.2)	9(2.4)	.892	.339	2.349	.817
	1-2 year	56(14.9)	33(8.8)	2.018	1.093	3.726	.025*
	3-5	92(24.5)	92(24.5)	1.189	.704	2.008	.517
	>5	37(9.9)	44(11.7)				1
Marital status of sellers	Single	39(10.4)	44(11.7)				1
	Married	136(36.3)	126(33.6)	1.337	.799	2.236	.269
	Divorced	11(2.9)	8(2.1)	1.976	.795	4.912	.142*
	Widowed	8(2.1)	3(0.8)	2.118	.751	5.968	.156*
Site of collection	Bore	76(20.3)	67(17.9)	1.403	.873	2.257	0.162*
	Hora Gibe	59(15.7)	41(10.9)	1.780	1.052	3.012	0.32
	Jiren	59(15.7)	75(19.5)				1
Type of market	Grocery	13(3.5)	19(5.1)				1
	Open market	181(48.3)	162(43.2)	1.633	.782	3.411	0.192*
Source	Farmers	116(30.9)	96(25.6)	.717	.472	1.088	0.117*

	Middle men	71(18.9)	82(21.9)	1.931	.486	7.670	0.350
	Private garden	7(1.9)	3(0.8)	1			
<i>Hygienic condition</i>	Adequate	7(1.9)	111(29.6)	1			
	Not adequate	187(49.9)	70(18.7)	42.361	18.813	95.383	<0.001*
<i>Type of product</i>	Vegetables	98(26.1)	89(23.7)	1.125	.740	1.710	.583
	Fruits	96(25.6)	92(24.5)				
<i>Cutaneous lesions</i>	Adequate	191(50.9)	179(47.7)				
	Not Adequate	3(0.8)	2(0.5)	.711	.117	4.307	0.771
<i>Discharge from nose, eye, ear and cough during visit</i>	Adequate	167(44.5)	177(47.2)	1			
	Not	27(7.2)	4(1.1)	7.154	2.451	20.881	<0.001*
<i>Wash status</i>	Adequate						
	Yes	57(15.2)	103(27.5)				
	No	137(36.5)	78(20.8)	3.174	2.072	4.862	<0.001*
<i>Means of display</i>	Floor	156(41.6)	132(35.2)	1.524	.940	2.470	0.087*
	Table/Shelf	38(10.1)	49(13.1)	1			
<i>Knowledge</i>	Good Knowledge	91(24.3)	119(31.7)	1			
	Poor Knowledge	103(27.5)	62(16.5)	2.172	1.432	3.296	<0.001*
<i>Attitude</i>	Positive attitude	90(24)	105(25)	1			
	Negative attitude	104(30.3)	76(27.7)	1.596	1.061	2.402	<0.001*
<i>Practice</i>	Good practice	131(34.9)	92(24.5)	1			
	Poor practice	63(16.8)	89(23.7)	2.012	1.324	3.057	0.001*

*Refers to candidate variables for multiple logistic regression

Finally, multiple logistic regression analysis was performed by putting all the thirteen variables which were candidate by bivariate analysis using SPSS by enter method and variables whose P-value is less than 0.05 were fitted in the final model. Among thirteen variables entered in multiple logistic regressions only four variables (fruits and vegetables washed before display, hygienic condition of the vender's/seller's environment, experience and study site) showed statistically significant association with bacterial contamination.

By putting all other variables constant the likelihood of bacterial contamination of fruits and vegetables is 0.088 times in fruits and vegetables bought from seller who had less than one year experience than those who had greater five years (95% C.I. 0.020 -.379 p-value=0.001), 3.538 times in fruits and vegetables bought from Hora gibe than Jiren (C.I. 1.529-8.185 P-value=0.003) 57.110 times in fruits and vegetables bought from sellers whose hygienic condition of the vender's/seller's environment is not adequate than adequate (C.I. 22.451-145.275 P-Value= <0.001) and 2.197 times in vegetables and fruits not washed before display than washed (C.I.1.121-4.309, P-value=0.022).

In table 9 below multiple logistic regressions output is presented including AOR with 95% C. I and P-value of all variables entered.

Table 9; multiple logistic regression output which shows factors associated with bacterial contamination

VARIABLE	CATEGORY	AOR	95% CI FOR COR		P VALUE
			Lower	Upper	
Educational Status Of Sellers	No formal education	.623	.221	1.762	.373
	Primary	.889	.425	1.863	.756
	Secondary and above				
Experience Of Sellers	<1 year	.088	.020	.379	.001*
	1-2 year	.476	.164	1.385	.173
	3-5	.649	.265	1.588	.344
	>5				1
Marital StatusOf Sellers	Single				1
	Married	1.278	.588	2.778	.535
	Divorced	1.862	.487	7.120	.364
	Widowed	2.769	.500	15.343	.244
Site Of Fruits Or Vegetables Collected	Bore	1.932	.985	3.790	0.055
	Hora Gibe	3.538	1.529	8.185	0.003*
	Jiren	1			
Type Of Market	Grocery				
	Open market	.526	.148	1.875	0.322
Source Of Vegetables And Fruits	Farmers	.236	.034	1.623	.142
	Middle men	.157	.022	1.134	.067
	Private garden				
Hygienic Condition of sellers	Adequate	1			
	Not adequate	57.110	22.451	145.275	<0.001*
Discharge From Nose, Eye, Ear And Cough during visit	Adequate	1			
	Not	3.057	.867	10.776	0.087

	Adequate				
Wash Status	Yes	1			
	No	2.197	1.121	4.309	0.022*
Means Display	On the floor	.676	.262	1.745	0.419
	On the table/shelf	1			
Knowledge	Good Knowledge	1			
	Poor Knowledge	1.101	.574	2.110	0.773
Attitude	Positive attitude	1			
	Negative attitude	1.113	.570	2.171	0.754
Practice	Good practice	1			
	Poor practice	.983	.486	1.991	0.963

Model fitness was tested with Hosmer and Lemeshow test and the assumption were fulfilled as the p-value of the Hosmer and Lemeshow was not significant.

Model fitness

Step	Chi-square	Df	Sig.
1	13.716	8	.089

4.4. Antimicrobial sensitivity and resistance pattern

Antimicrobial susceptibility testing (AST) for isolated pathogens was performed by disk diffusion method according to Clinical Laboratory Standards Institute (CLSI, 2021) guidelines to the following antimicrobial agents on Mueller–Hinton agar (Oxoid): Ciprofloxacin (CIP-5µg), Gentamycin (GM-10µg), Ceftriaxone (CRO-30µg), Piperacillin-tazobactam(TZP-100/10µg), Erythromycin (EM-15 µg), Trimethoprim-sulfamethoxazole (co-trimoxazole) (SXT-1.25/23.75 µg), Meropenem (MEM-10µg), Ampicillin (AMP- 10µg), and cefazolin(CZN-30µg), Tobramycin(TOB-10µg), Cefoxitin (FOX-30µg), Penicillin(PCN-10µg),Clindamycin(CLI-2µg), Amoxicillin-clavulanate(AMC-20/10µg). Almost all E-coli spp isolated were sensitive to Piperacillin-tazobactam (95.3%), cefazoline (93%) and cotrimoxazole (93%), ciprofloxacin (83.6%) and ceftriaxone (76.7%), while majority of the species were resistant to amoxicillin-clavunic acid. Majority (86.8%) of Klebsiella spp isolated were susceptible to Cotrimoxazole (Table 10).

Table 10: Percentage of Resistance of isolated Bacteria species

S · N	Isolated Gram negative and Gram Positive Bacteria species	CIP-5µg)	GM-10µg	CRO-30µg	TZP-100/10µg	EM-15 µg),	SXT-1.25/23.75µg)	MEM-10µg	AMP- 10µg	CZN-30µg	(TOB-10µg	FOX-30µg	PCN-10µg	CLI-2µg	AMC-20/10µg
1	E-coli Spp (n=43)	16.3	39.5	23.3	4.7	NA	7	15	NA	7	44.2	NA	NA	NA	90.7
2	Klebsiella Spp (n=38)	39.5	63.1	39.5	15.8	NA	13.2	25	NA	39.5	76.3	NA	NA	NA	92.1
3	Salmonella Spp (n=66)	30.3	19.7	31.8	NA	NA	3	NA	98.5	NA	NA	NA	NA	NA	80.3
4	Shigella Spp (n=15)	26.7	60	60	NA	NA	20	NA	100	NA	NA	NA	NA	NA	33.3
5	S. aureus (n=77)	2.6	NA	NA	NA	62.3	2.6	NA	NA	NA	NA	7.8	84.4	5.2	NA

Both salmonella and Shigela species showed highest susceptibility for cotrimoxazole 97% and 80% respectively. 98.5% of salmonella species and 100% Shigela species develop resistance for ampicillin (Table 10).

Isolated *S. aureus* species were susceptible for most of antibiotics tested, ciprofloxacin (97.4%), cotrimoxazole (97.4%), clindamycin (94.8%) and cefoxitin (92.2%). But resistance was observed for penicillin (84.4%) and erythromycin (62.3%) (table10).

Generally, Ciprofloxacin was the most effective drug (79.9%) followed by Cefazoline (77.7%), cotrimoxazole (74.58%), ceftriaxone (66.4%), and Gentamicin (61.15%). But Highest resistance was observed in penicillin (84.4%) followed by ampicillin (81.4%).

5.5 Knowledge, Attitude and practice towards food safety (IP and bacterial contamination of vegetables and fruits) of vendors

The present study assessed the level of vendor’s knowledge, attitude and practice towards about intestinal parasitic and bacterial contamination of vegetables and fruits using standardized Likert scale questions among 375 fruit and vegetable sellers/vendors. And participant’s response for each item was summated and classified based on the mean score of respondents.

Respondents KAP were classified based the mean score. Accordingly, 152(40.3 %), 210(55.2%) and 195(52.0%) of the respondents had good practice, good knowledge and positive attitude towards food hygiene and safety respectively (Table13).

Table 11: KAP of vendors towards fruits and vegetables hygiene and safety (n=375)

Variable	Category	Frequency	Percent
Attitude	Positive Attitude	195	52.0
	Negative Attitude	180	48.0
Knowledge	good knowledge	210	56.0
	poor knowledge	165	44.0
Practice	poor practice	223	59.5
	good practice	152	40.5
Total		375	100.0

CHAPTER SIX

6. DISCUSSION

6.1 Parasitic contamination

In the present study, a total of 375 Sellers were participated. The mean age of the participants was 29.02 ± 4.99 years and 91.2% of them were females. A total of 375 vegetable and fruit samples were analyzed and 173 samples were found contaminated with at least one type of parasite. From the total 375 samples, 187(49.9%) were vegetables and the rest 188 (50.1%) were fruits. *A.lumbricoides* 46(12.3%) was the most frequently detected parasite, followed by *Strongyloidesstercoralis* 32 (8.5%).

In this study, the prevalence of intestinal parasites was 46.1%. This finding was lower than findings of similar studies in Brazil (50.9%); Yemen (100%); Cape Coast metropolis market (52.4%), and Koforidua (57.5%) in the Eastern Region of Ghana; Jos State in Nigeria (56.25%); Asmara State of Eritrea (57.07%); Dire Dawa (47.3%), Arba Minch (54.4%), Jimma (57.8%) and Dessie (63.4%) towns in Ethiopia(6,10,26,55–61).The difference could be due to variations in items of samples collected, processing and laboratory methods used. For example, in the study from Ghana (57), fruits and vegetables were thoroughly washed twice with saline in order to increase recovery of parasites; but in the present study, samples were washed only once. In the study from Brazil, vegetable samples (lettuce, green onion, and salad) were collected between February and July and triplicate slides were examined (55). All these features contribute to a higher contamination rate than in the present study. Time of sample collection might be another factor in those studies in Dire Dawa(56), Arba Minch (6), Jimma(10), and Dessie(59) towns in Ethiopia were conducted before the launching of the national mass drug administration and water, sanitation and hygiene (WASH) programs in the country. Since 2015, many WASH activities, health education, and biannual deworming programs for helminths have been conducted and substantially decreased environmental fecal contamination as well as parasitic contamination. The parasitic contamination rate in Arba Minch town was reduced from 54.4% in 2014 to 25.1% in 2018 (6,7) Variations in geographical distribution of parasites, sanitary and socioeconomic status of the community could be the other contributing factors. Alternatively, the overall contamination rate in the present study was higher than findings in Iran (62) (8.4%); Ibadan city of Southwest Nigeria (11.6%) (40); Benha (29.6%) in Egypt (11) ;

Sudan (63) (10.6%) and Arba Minch(7) (25.1%) and Bahir Dar (64) (39.1%) towns in Ethiopia. Factors mentioned above could also contribute here. For instance, in the study from Egypt, samples were washed with tap water for 6–7 min for removal of mud and dust before being immersed in physiological saline. This could decrease as some of the parasites might be removed with mud and dust particles. A study from Sudan screened only 150 samples and non-leafy items were screened and samples were not processed by modified acid-fast staining; therefore, oocysts of intestinal coccidian were not assessed; all contributing to a lower contamination rate compared to the present study. In a study from Arba Minch, a variable number of fruit and vegetable samples from each item were screened unlike the equal number of each item in the present study. This might bring variation in the two studies because there is a difference in susceptibility to contamination among different items of fruits and vegetables (7,11,63).

The current finding showed that the highest prevalence of parasites was detected in carrot 30 (63.8%), followed by lettuce 29(63.1%) and Cabbage 29(61.7%). This might be due to the fact that edible parts of vegetables grow under or closer to the soil than that of fruits. Hence, soil may also play significant role in the contamination of vegetables (28,65) Banana with 7(14.9%) prevalence is the least contaminated fruit in the present study. Parasitic contamination was 5.78 times in vegetables compared to fruits in the present study which is in line with the study conducted at Bahir Dar city (29). And in addition to the above-mentioned reason this could be due to roughness of surfaces of vegetables which facilitates attachment of parasites and even it will help the parasites to resist easy washing a compared to smooth surface of fruits (66).

A. lumbricoides was the most frequently detected parasite in the present study with prevalence of 12.3%. This finding is consistent with previous studies conducted in Ethiopia and elsewhere (6,10,13,66–70). This might be due to parasite's cosmopolitan nature, the high number of eggs produced by the female parasite, and the strong and resistant nature of the eggs that enable them to survive harsh environment. It is known that the ova can survive in the absence of oxygen, live for 2 years at 5–10°C, and be unaffected by desiccation for up to 3 weeks (71).

The finding of the present study revealed that washing of fruits and vegetables before display was the most significant factor contributing parasitic contamination (decrease/Increase). This finding is in agreement with similar previous study in the city and other studies conducted at

Bahir Dar, Dire Dewa and Arba Minch towns in Ethiopia (6,10,56,64). This can be justified by the fact that washing before display will remove parasites. But the finding of the present study is in contradiction with another study conducted at Aksum town in Ethiopia which reported washing before display will increase parasitic contamination of fruits and vegetables. This variation might be due to contamination of water used for washing of fruits and vegetables and washing process difference of venders or sellers.

Another important contributor of parasitic contamination found in the current study was means of display of fruits and vegetables. The likely hood of parasitic contamination was 54.766 times higher in fruits and vegetables displayed on the floor than in fruits and vegetables displayed on the table/shelf (P-value= <0.001). This finding is in line with a study conducted Dire Dewa (56). This is might be due to the fact that fruits and vegetables displayed on the floor are exposed to dusts and flies as there is established fact flies can act as a vector for parasites. The present study revealed that the odds of parasitic contamination of vegetables and fruits was 21.7 times higher in fruits and vegetables which was bought from venders or sellers who had no formal education than who had attended at least secondary school education. This finding is consistent with a study conducted at Aksum and Tarcha towns in Ethiopia (13,72).

6.2 Bacterial Contamination of Fruits and Vegetables

In addition to parasitic contamination, the present study attempted to see bacterial contamination of fruits and vegetables. As a result, it was found that overall bacterial contamination rate of 51.7%. This finding is lower than other studies of its kind from Bangladesh and India where prevalence of bacterial contamination were 97.3% and 100% respectively (73,74). However, it is higher than a study conducted at Arba Minch town, Ethiopia (75) which reported 48.7% prevalence, USA(76) and Canada (77).

Among nine species of bacteria detected in the present study, *S. aureus* was the most prevalent one with 77(20.5%) prevalence. This is in line with the study conducted at Cameroon, Nigeria, Amravati City in India, Bangladesh and Sudan where *S. aureus* was the most dominant bacterial contaminant (73,78,78,79). In contrary, previous study from Jimma town reported that *S. aureus* was the least contaminant of fruits and vegetables while reporting *Enterobacter spp.* (21.60%) was the highest contaminant (80). This variation could be due to the difference in sampling/collection of fruits and vegetables. The prevalence of *E-coli* in the present study was

44 (11.5%) which was lower compared to previous studies (75,80). These differences might be due to variations in the time (season) of data collection and sensitivity of laboratory methods used. In the present study, avocado was the most frequently contaminated fruits (44, 93.6%). But reports from previous study showed that cabbage had highest contamination rate (75).

The present study also sought to see factors contributing bacterial contamination of fruits and vegetables. Accordingly, the most important factor was hygienic condition of seller's environment where fruits and vegetables bought from sellers whose environmental hygiene is not adequate had 57.1 times risk of bacterial contamination.

Other contributing factor was wash status of vegetables and fruits as it was found that the odds of bacterial contamination was 2.197 times in vegetables and fruits that are not washed before display than washed in the present study. This finding is supported by another study from Ghana (30) and Nekemte town, Ethiopia (81) which reported washing of vegetables and fruits will reduce bacterial load of fruits and vegetables. However, other researchers argue that washing of fresh fruits and vegetables may prove ineffective to remove microorganisms, as they may remain attached to surfaces of fruits and vegetables (82), or become internalized in the edible parts of the fruits and vegetables (83) and thus, are not accessible for efficient removal.

In the present study, the odds of bacterial contamination was 3.538 times in fruits and vegetables bought from Hora gibe than Jiren and this finding is in line with a study conducted at Arba Minch town, Ethiopia which reported Vegetables sold in Sikela and Shecha markets were 56.4% and 85.7% less likely to be contaminated by bacteria respectively compared to vegetables sold in Konso sefer (75). This might be due to the way vegetables are handled differs among markets so that the tendency to be contaminated also varies.

Isolated *S. aureus* species were susceptible for ciprofloxacin (97.4%), cotrimoxazole (97.4%), clindamycin (94.8%) and cefoxitin (92.2%). But resistance was observed for penicillin (84.4%) and erythromycin (62.3%).

In the current study, Salmonella species showed susceptibility for cotrimoxazole (97%) followed by gentamicin (80.3%). But 98.5% and 80.3% of salmonella species develop resistance for ampicillin and Amoxicillin plus clavunic acid respectively. This finding is fairly in line with previous study in the area which reported 100% and 93% of salmonella spp. was

susceptible gentamycin and co-trimoxazole respectively (80) . Salmonella spp. susceptibility to gentamicin was also in line with a study by Akbarmehr (84)who reported that Salmonella spp. were highly susceptible to chloramphenicol (100%) followed by gentamycin (91.89%). The antibiotic resistance patterns of *S. aureus* isolates in the current study showed low percentage of resistance to Ciprofloxacinand Cotrimoxazole (2.6%for each) This is partly similar to previous report from Ghana (85). In the current study, *S. aureus* isolated were resistant to penicillin G (84.4 %), and Erythromycin (62.3%) This finding is partly in agreement with Sina et al. (85).

6.3 limitation of the study

Lack of measure the intensity (colony count) of contamination.

Prevalence of parasitic contamination might be underestimated due the utilization of light microscope due to lack of florescent microscope.

CHAPTER SEVEN

7. CONCLUSIONS AND RECOMMENDATIONS

7.1 CONCLUSIONS

- The level of parasitic and bacterial contamination of fruits and vegetables sold at peri urban kebeles of Jimma town was high.
- Vegetables are more prone to parasitic contamination than fruits.
- Besides parasitic contaminants, nine species of bacteria were detected *S. aureus* being the commonest bacterial contaminant of fruits and vegetables
- Majority of the vendors had poor practice of handling fruit and vegetable.
- Ciprofloxacin was the most effective drug for isolated bacteria.
- Fruits and vegetables not washed before display, unhygienic condition of the vendors/seller's environment, experience and study site) were identified factors for parasitic contamination.
- Similarly, factors contributing for bacterial contamination were experience of sellers, fruits and vegetables not washed before display, in adequate environmental hygiene and site where vegetables and fruits were bought.

7.2 Recommendations

- Since the contamination of fruits and vegetables by pathogenic intestinal parasites and bacteria, vendors should improve their handling practices starting from transportation to display to the customers.
- Though average number of respondents had good knowledge about fruit and vegetable handling, still there should be comprehensive health education to vendors.
- Vendor should use shelf or table rather than displaying their fruits and vegetables in floor.
- Professionals and other people including farmers and vendors involved in the vegetable and fruit production from production to the market should be made aware of the potential risk associated with various practices and possible chances of contamination.

They should be educated to gain sufficient knowledge on the source of etiological agents responsible for the contamination and their resultant diseases

- Continuous supervision of vendors should be done by the public health sectors to improve handling practice of vendors.
- Periodic on-market screening of fruits and vegetables should be done.
- Local and regional concerned bodies should regulate and control vendors hygiene and safety practice.
- Attention should be given to rational use of drugs to prevent microbial resistance.
- Further studies should be conducted on the viability of parasitic and bacterial contaminants of fruits and vegetables using strong study designs.

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ANNEX I: INFORMATION SHEETS

1.1. Information sheet English version

This information sheet was prepared for individuals who were volunteer to participate in the study. The detailed explanation about what was undertaken in the study was presented as follows and it was after reading the description that informed consent was obtained.

Title of the project: Intestinal parasitic and bacterial contamination of vegetables and fruits sold in open-aired markets in selected peri-urban kebeles of, Jimma town, southwest Ethiopia

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Description and Purpose of the study: -Determining of Intestinal parasitic and bacterial contamination of vegetables and fruits sold in open-aired markets important to give appropriate measurement. Because raw fruits and vegetables as the potential source of transmission for intestinal parasites and bacteria to humans. Foodborne intestinal parasitic and bacterial infection can be reduced most effectively by preventing contamination of fruits and vegetables. The result of this study will provide useful information to the role of concerned body in the control of parasitic and bacterial contamination in these vegetables and fruits and also will be used as a base line for those who are interested in carrying out further study. Moreover, this study provides information for the Jimma town health office administrators. Hence this study will be conducted to determine prevalence of Intestinal parasitic and bacterial contamination of vegetables and fruits sold in open-aired markets of peri-urban of JimmaTown.

Procedures: -Following your willingness you are asked to sign a written consent and the following procedures will be undertaken

- You will provide us 15 minutes for interview

- Fruits and Vegetables samples will be collected /purchased.
- For parasitic identification, the sediment will be examined under a light microscope and modified Zeihl-Neelsen staining technique will be used for identification of oocysts
- For bacterial identification, vegetables and fruits will be washed by sterile distilled water and prepared serial dilutions will be cultured using culture media.
- Disc diffusion techniques are used to determine the drug sensitivity of the isolated pathogenic bacteria.

Risks and discomforts: -During sample collection/purchasing there is no risks and discomforts.

Benefits: - This study will be of benefit to the entire community. There is no direct financial benefit you get by participating in this study but the study result will be delivered timely and appropriate intervention will be pointed.

Confidentiality: -Any information obtained during this study will be kept confidential. This is assured by avoiding use of any identifier and information will be recorded with code number.

Voluntary participation: - Participation on this study is voluntary and you have the right to refuse participation at any time. Your decision will not result in any penalty or loss of benefits to which you are entitled. Your decision will not put you at risk.

You may ask questions now and, in the future, if you do not understand something that is being done contact the investigator on above address.

For the success of the study, I will be asking you to give the correct answer for the respective questions. Thank you for your assistance!

Signature of participant: _____

Date: _____

1.3. Ibsa Hirmaattota qo'annootif guca guutamu (Afaan Oromoo)

Guciiin Kun Kan guutamu warren qo'annaa irratti fedhiin hirmaataniif Kan ooluu fi haallii qo'annaa sirritti erga ibsameefii booda Kan guutamu fi Kan mallattaa'udha

Mata-duree qo'annaa:-Qorannoon Magaala Jimmaatti Gandoota Filataman sadan(Jireen,Booree fi Hora Gibee) irratti Maxantuulee garaa fi Baakteriyaa Faalama Kuduraa fi muduraa Gabaaratti gurguraman fidan qorachuudhaaf kan fayyadamnu.

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Dhimmi-qo'annichaa: Faalama kuduraa fi fuduraa gabaatti bakke gurguraman beekuun barbaachisaa dha. Sababiin isaas faalamni fuduraa fi kuduraa tattamsa'ina Maxantoota garaa fi baakteeriyaa sababa isa guddaa dha.Faalama kuduraa fi fuduraa ittisuun dhukkubootaa maxantoota garaa fi baakteeriyaa darban ittisuuf ni fayyada.Kanaafuu bu'aan qorannoo kana qaamolee ittisa dhukkuboota daddarboo irratti hojetaniif madda odeeffannoo ta'uu waan danda'uuf qorannoon kun barbaachisaa dhaHaala adeemsa qo'annichaa: - qo'annaa irratti fedhiin hirmaachuu keessan Mallattoo keessaniin nuuf ibsitaaniif ragaalee armaan gadii kanneen nuuf kennitan.

Gaaffiilee afaaniitiif daqiiqaa 15 waliin turra

Kuduraa fi fuduraa muraasa ni fudhanna

Kuduraa fi fuduraa qorannoo maxantuu garaa fi Baakteeriyaa ni qoratama

Wantootni armaan olii Kun qorannoof Kan barbaachisan tahu ni ibsina.

Sodaa fi miidhaa qabu: -seeraa fi naamusa ogummaa fayyaa hordofuun waan dalagamuuf wanti nama sodaachisu hin jiru

Faayida qo'anniichaa fi kafaaltii hirmaataaf godhamu- qorannoo irratti hirmaachuuf kafaltii kan hin qabnee fi bu'a qorannoo irraa argamuu faayidaa keessanii fi ummataa ta'uu hubachuu qabdu.

Iccitii qo'annichaa-wantootni qorannoo irraa argaman hundinuu icciitiin kan eeggamaniifi ragaaleen argaman hundinuu maqaa keessaniin osoo hin tahiin lakkoofsa addaatiin kan beekkamaniifi odeeffannoon hundinuu iccitiidhan warra ragaa funaanan biratti kan hafu tahuu isaa isiiniif ibsina.

Mirga fedhaan hirmaachuu- qorannoo irratti hirmaachuun fedhii kee qofa tahuu isaa beektee, yeroo barbaadetti qorannoo keessaa bahuu kan dandeessu fi yeroo keessaa baatulee rakkoo tokkoo kan sirratti hin fidnee fi tajaajila argachuu qabdu hundumaa argachuu kan dandeessu tahuusaa fi qorannoof hirmaachuu keetiif galannii keenya guddaadha.

Galatoomi!!

ANNEX: CONSENT /ASSENT/ FORMS

2.1. Consent/Assent forms (English version)

Participant Code Number _____

Study participants will be informed about required fruits and vegetables for examination of intestinal parasites and bacterial contamination of fruits and vegetables sold in open aired markets. Consequently, after seller agreed, the questioner will be asked and fruits and vegetables will be purchased. I am informed fully in the language I understand about the aim of the above-mentioned research. I understood the purpose of the study entitled with “intestinal parasites and bacterial contamination of fruits and vegetables sold in open aired markets in selected peri urban kebeles of Jimma Town. In addition, I have been told all the information collected throughout the research process will be kept confidential. The secrecy of information by anonymity. I have the right to left from the research at any time without in any way affecting. I have given my consent freely to participate in the study.

Agree _____ Not agree _____

Therefore, I give my consent without any coercion for my participation in this study.

Participant's full Name _____ signature _____ Date _____

Investigator's name _____ signature _____ Date _____

Witness 1. Name _____ signature _____ date _____

2. Name _____ signature _____ date _____

2.2. የስምምነት ቅጽ በአሚኛ

የተሳታፊው ስም _____

የተሳታፊው አድራሻ _____

የጥናቱ ተሳታፊ በገላጣግ በያዎቸው ውስጥ በሚሸጡ አትክልቶች እና ፍራፍሬዎች ጥገኛ እና ባክቴሪያ ብክለትን ለመመርመር መርመራዎቻችን እና ፍራፍሬ እና አትክልት ምርመራ እንደሚደረግ እና ምንም አይነት እንደሌለው ይነገራቸዋል፡፡ ከላይ ስለተጠቀሰው የምርመራ ዓላማ በተረዳሁት ቋንቋ ማረጃ ተሰጥቶኛል፡፡ በደቡብ ምዕራብ ኢትዮጵያ በጅም ከተማ በተመረጡ የከተማ ቀበሌዎች ውስጥ በገላጣግ በያዎቸው ውስጥ በሚሸጡ አትክልቶች እና ፍራፍሬዎች ጥገኛ እና ባክቴሪያ ብክለትን ለመመርመር መርመራ ርዕስን የጥናቱን ዓላማ ተረድቻለሁ፡፡ የአትክልቶች እና ፍራፍሬዎች ምርመራ እንደሚደረግ እና በጥናቱ ማሳተፍ ወቅት ምንም ስጋት እንደማይኖር ተነግሮኛል፡፡ በተጨማሪም በጥናቱ ሂደት በሚሉ የተሰበሰበው መረጃ ሁሉ በሚጠበቅ እንደሚሆን ተነግሮኛል፡፡ በማንኛውም ጊዜ ከምርመራ የመውጣት መብት አለኝ፡፡

በጥናቱ ላይ ለመሳተፍ በነፃነት ፈቃዴን ሰጥቻለሁ፡፡
እስማሙ _____ አልስማም _____
ስለሆነ ምክብር ጥናት ውስጥ ለመሳተፍ ለምንም ስጋት ለማይኖር ፈቃዴን እሰጣለሁ፡፡

የተሳታፊው ፊርማ _____

የዋና ተመራማሪ ፊርማ _____

ምሳክር

1. ስም _____ ፊርማ _____ ቀን _____

2. ስም _____ ፊርማ _____ ቀን _____

2.3. UunkaaWalii galtee (Afaan Oromoo)

Lakk.addaa Hirmaattota_____

Maqaan guutuu Hirmaattota_____

Ani hirmaatan maqaan koo armaan olitti ibsame kun bu'aa fi miidhaan isaa erga sirritti natti himame fi miidhaan omaa kan hin qabne ta'uu isaa ergan hubadhee, booda, saamuda qorannoon laboratoritiif kan oolu fuduraa fi kuduraa koo kennuu fi Dabalataaniis odeefannoo narraa argaman hunduu icciitiin akka qabaman nattii hiimameera. Akkasumas gaaffileen gaafatamuuf deebii kennuu dhiisuu, hiirmachuu dhiisuu fi yeroon barbaadetti addaan kutuu akkan danda'uu bareen jira.Kana godhuu kiyyaaniis ammas ta'ee fuulduraaf fayyadamummaa tajaajila fayyaa kiyya irratti rakkoon tokkollee akka hin uumamanee huubadheen jira.

Walii galeera_____ **walii hin gallee**_____

Kanaafuu qorannoo kana irratti fedhiin hirmaachuu kiyya Mallattoo kootiin nan mirkaneessa.

Maqaa hirmaataa_____Mallattoo_____Guyyaa_____

Maqaa qorataa_____Mallattoo_____Guyyaa_____

Ragaalee

1. Maqaa_____Mallattoo_____Guyyaa_____

2. Maqaa_____Mallattoo_____Guyyaa_____

Annex-III: Data collection tool

Table: 4. Questionnaires for assessment of parasitic and bacterial contamination of vegetables and fruits and for assessment of KAP of sellers' about parasitic and bacterial contamination of vegetables and fruits sold in open-aired markets in selected peri-urban kebeles of, Jimma town, southwest Ethiopia

A. Sociodemographic variables and their association of fruits and vegetables sellers'

S No.	Question	Response
1	Sample code	_____
2	Gender	1.male 2.Female
3	Age of respondent	_____years old
4	Date of collection	_____
5	Study site	_____
6	Market name	
7	Market type	1. Grocery 2. Open market
8	*Type of vegetable/fruit	_____
9	Source of vegetable/fruit	1. Farmers 2. Middle men 3. Private garden
10	Means of vegetable display	1. On the floor 2. On shelf in shop 3. On tables 4. On wheel barrow
11	Vegetable washed before display?	1. Yes 2. No
12	What type of water source do you use for your home	1. Pipe water 2. Well water

	(washing vegetables and fruits)	3. River water
13	What is your marital status	1. Single 3. Divorced 2. Married 4. Widowed
14	Educational status of the seller	1.No formal education 2.Primary education 3.Secondary education 4. Higher education
15	Monthly Income (in Ethiopiab Birr)	_____
16	Fruits and vegetables safety Training	2. No 2. YES
17	Experience (years) of vendors	1. < 1 3. 3-5 2. 1-2 4. > 5

B. Fruits and vegetables safety knowledge assessment of Fruits and vegetables vendors’/Sellers’

1.	Bacteria and intestinal parasites can be contaminated fresh fruits and vegetables.	2. True 2.False 3.Do Not Know/Do Not Remember
2.	Contaminations of fresh fruits and vegetables may happen internally or externally during farming, harvest, stuffing, storage, transporting, marketing and shops, and even at the consumer's home.	1.True 2.False 3.Do Not Know/Do Not Remember
3.	Proper Washing and cooking fruits and vegetables before conception is the possible prevention for bacteria and intestinal parasites contamination of fruits and vegetables	1.True 2.False 3.Do Not Know/Do Not Remember
4.	Washing hands before work reduces the risk of Fruits and vegetables contamination	1.True 2.False 3.Do Not Know/Do Not Remember
5.	A healthy Fruits and vegetables vendors’ may contaminate food with microbes that causes foodborne diseases.	1.True 2.False 3.Do Not Know/Do Not Remember

6	Fruits and vegetables vendors' health status must be periodically checked.	1.True 2.False 3.Do Not Know/Do Not Remember
7	Washing fruit and vegetables under running water and peeling them is not enough to make these foods safe for consumption.	1.True 2.False 3.Do Not Know/Do Not Remember
8	Well cooked vegetables are free from microbes that cause foodborne diseases.	1.True 2.False 3.Do Not Know/Do Not Remember
9	Fruits and vegetables vendors with cuts or wounds on hands need to be kept away from Fruits and vegetables handling activities	1.True 2.False 3.Do Not Know/Do Not Remember
10	The consumption of Raw vegetables and fruits are can be cause of disease.	1.True 2.False 3.Do Not Know/Do Not Remember
11	Lack of hygiene of vegetables and fruits is the cause of intestinal parasitic infections and bacterial infections	1.True 2.False 3.Do Not Know/Do Not Remember
12	Inappropriately waste disposed around your working area/markets can be source of microorganisms that will contaminate fruits and vegetables	1.True 2.False 3.Do Not Know/Do Not Remember
13	Consuming/eating raw/uncooked fruits and vegetables has not only benefits but also risks/harms.	1.True 2.False 3.Do Not Know/Do Not Remember

C.Fruits and vegetables safety attitudes assessment of Fruits and vegetables vendors

	Question	Responses
1	Frequent hand washing is worth the Time	1. Extremely Agree 2. Agree 3. Neutral 4. Disagree 5.Extremely Disagree
2	Improper Fruits and vegetables storage may pose risk to health.	1. Extremely Agree 2. Agree 3. Neutral 4. Disagree 5.Extremely Disagree
3	Keeping surfaces clean reduces risk of fresh products contamination	1. Extremely Agree 2. Agree 3. Neutral 4. Disagree 5.Extremely Disagree

4	Washing hands before handling raw fruits and vegetables reduces the risk of fresh products contamination	1. Extremely Agree 2. Agree 3. Neutral 4.Disagree 5.Extremely Disagree
5	Fresh products vendors who have wounded fingers and hands can handle fruits and vegetables only if they correctly cover their cuts.	1. Extremely Agree 2. Agree 3. Neutral 4.Disagree 5.Extremely Disagree
6	Safe food (fruits and vegetables) handling is an important part of my job responsibilities	1. Extremely Agree 2. Agree 3. Neutral 4.Disagree 5.Extremely Disagree
7	Health education can reduce the prevalence of intestinal parasitic and bacterial infections	1. Extremely Agree 2. Agree 3. Neutral 4.Disagree 5.Extremely Disagree
8	It is important to wash hands right after unhygienic practices	1. Extremely Agree 2. Agree 3. Neutral 4.Disagree 5.Extremely Disagree
9	Uses of soap while washing hand can prevent intestinal parasitic infections and bacterial infections	1. Extremely Agree 2. Agree 3. Neutral 4.Disagree 5.Extremely Disagree

D.Fruits and vegetables safety practices assessment of Fruits and vegetables Vendors

	Question	Responses
1.	Do you wash your hands immediately before handling Fruits and vegetables?	1.Never 2.Rarely 3.Sometimes 4.Most of the Times 5.Always
2	Do you use the toilet for defecation?	1.Never 2.Rarely 3.Sometimes 4.Most of the Times 5.Always
3	Do you wash your hand after toilet with soap/ash?	1.Never 2.Rarely 3.Sometimes 4.Most of the Times 5.Always
4	Do you handleFruits and vegetables when you are sick or have cuts on hands?	1.Never 2.Rarely 3.Sometimes 4.Most of the Times 5.Always
5	Do you cut your finger nails regularly?	1.Never 2.Rarely 3.Sometimes 4.Most of the Times 5.Always
6	Do you keep your hair completely covered with a cap while handling fruits and vegetables?	1.Never 2.Rarely 3.Sometimes 4.Most of the Times 5.Always
7	Do you sanitize/clean your workplace before work and after finishing your service?	1.Never 2.Rarely 3.Sometimes 4.Most of the Times 5.Always

8	Do you wash hands after rubbing your nose or scratching your body?	1.Never 2.Rarely 3.Sometimes 4.Most of the Times 5.Always
9	Do you wash fruits and vegetables before sale	1.Never 2.Rarely 3.Sometimes 4.Most of the Times 5.Always

E. The observations will be carried out with Fruits and Vegetables Vendors by observation checklist

Observed fruits and vegetables Safety Practice		Adequate / Inadequate
1	The personal hygiene of vendors includes the use of clean cloth,Nails are clean, and short	1. Adequate 2. Inadequate
2.	Vendors with cutaneous lesions and wounds or symptoms of diseases/infections	1. Adequate 2. Inadequate
3.	Discharging from nose, eyes, ear and cough during visit	1. Adequate 2. Inadequate
4.	Cleanness of the fresh products'(fruits and vegetables) container	1. Adequate 2. Inadequate

15	ወርሃዊ ገቢ (በኢትዮጵያ ብር)	-----
16	ፍራፍሬዎች እና አትክልቶች ደህንነት ስልጠና ወስደዋል	1. የለም 2. አዎ
17	የሻጮች ልምድ (ዓመታት)	-----

ለ. የፍራፍሬ እና የአትክልቶች ሻጮች? የፍራፍሬ እና የአትክልቶች ደህንነት ዕውቀት ግምገማ

1.	ከሥራ በፊት እጅን ማታጠጠ ፍራፍሬዎችን እና አትክልቶችን የማከል አደጋን ይቀንሳል	1. እወሳት 2. ሐሰት 3. አላውቅም / አላስታውስም
2.	ጓንት ማድረግ እጅን ለማጭዳት ምክንያት ነው :	1. እወሳት 2. ሐሰት 3. አላውቅም / አላስታውስም
3.	ጤናማ የፍራፍሬ እና የአትክልት ሻጮች ነበረው ወይስ በሽታዎች ምክንያት በመሞገጥ ማይክሮቦች አማካኝነት ምግብ ሊበክሉ ይችላሉ: :	1. እወሳት 2. ሐሰት 3. አላውቅም / አላስታውስም
4.	የአትክልት እና ፍራፍሬ ሻጮች የጤና ሁኔታ በየጊዜው ማረጋገጥ አለበት::	1. እወሳት 2. ሐሰት 3. አላውቅም / አላስታውስም
5.	ፍራፍሬዎችን እና አትክልቶችን በሚፈጸሙ ወሃ ማጠጠ እና እነሱን ማለቀቁ እነዚህን ምግቦች ለምግብነት ጤናማ ለማድረግ በቂ ነው :	1. እወሳት 2. ሐሰት 3. አላውቅም / አላስታውስም
6.	በደንብ የበሰሉ አትክልቶች ለምግብ ወለድ በሽታዎች ከማይስከት ማይክሮቦች ነፃ ናቸው :	1. እወሳት 2. ሐሰት 3. አላውቅም / አላስታውስም
7.	በእጅቻቸው ላይ ቁስሎች ያሉባቸው ፍራፍሬዎች እና አትክልቶች ሻጮች ከፍራፍሬ እና ከአትክልቶች አያያዝ ተግባራት ማቅ አያስፈልጋቸውም :	1. እወሳት 2. ሐሰት 3. አላውቅም / አላስታውስም
8.	ጥሬ አትክልቶች እና ፍራፍሬዎች ማጠቀሚያው ለበሽታ ማስኬ ሊሆኑ ይችላሉ: :	1. እወሳት 2. ሐሰት 3. አላውቅም / አላስታውስም
9.	የአትክልት እና ፍራፍሬ ንፅህና ጉድለት ለአንጀት ጥገኛ ኢንፎክሽኖች እና የባክቴሪያ ኢንፎክሽኖች ማስኬ ነው	1. እወሳት 2. ሐሰት 3. አላውቅም / አላስታውስም
10.	በቆዳው ተላላፊ በሽታ በተያዙ ወቅት ከሥራ ማቅ አስፈላጊ ነው :	1. እወሳት 2. ሐሰት 3. አላውቅም / አላስታውስም
11.	በሥራ በታዎ / በገቢያዎ ዙሪያ አግባብ ባልሆነ ማንገድ የሚገኙ ቆሻሻዎች ፍራፍሬዎችን እና አትክልቶችን የሚበክሉ ረቂቅ ተሕዋስያን ምንጭ ሊሆኑ ይችላሉ	1. እወሳት 2. ሐሰት 3. አላውቅም / አላስታውስም

12	ጥሬ / ያልበሰሉ ፍራፍሬዎችን እና አትክልቶችን / መጣጣብ ጥቅሞች ብቻ ሳይሆን አደጋዎች / ጉዳቶችም አሉት፡፡	1. እውነት 2. ሐሰት 3. አላወቅም / አላስታወስም
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ሐ / ፍራፍሬዎች እና አትክልቶች ሻጮች የፍራፍሬ እና የአትክልቶች ደህንነት አመለካከት ግምገማ

ተ.ቁ	ጥያቄ	ምላሾች
1	ተደጋጋሚ የእጅ መታጠብ ዋጋ አለው	1. እጅግ በጣም አልስመናም 2. አልስመናም 3. ገለልተኛ 4. እስመናላሁ 5. በጣም እስመናላሁ
2	ተገቢ ያልሆኑ ፍራፍሬዎች እና አትክልቶች መክመቶ ለጠጃነት አስጊ ሊሆኑ ይችላሉ፡፡	1. እጅግ በጣም አልስመናም 2. አልስመናም 3. ገለልተኛ 4. እስመናላሁ 5. በጣም እስመናላሁ
3	ንጥፎችን በንጽህና መጠበቅ የንጹህ ምርቶች ብክለት አደጋን ይቀንሳል።	1. እጅግ በጣም አልስመናም 2. አልስመናም 3. ገለልተኛ 4. እስመናላሁ 5. በጣም እስመናላሁ
4	መደበኛ የምግብ መከከል ቁጥጥር አሰራርን በትክክል መከናወን በቂ መከላከያ ይሰጣል ለተጠቃሚዎች የኢንፎክሽን መከተላለፍን ይከላከላል፡፡	1. እጅግ በጣም አልስመናም 2. አልስመናም 3. ገለልተኛ 4. እስመናላሁ 5. በጣም እስመናላሁ
5	ጥሬ ፍራፍሬዎችን እና አትክልቶችን ከመያዝ በፊት እጅን መታጠብ ትኩስ ምርቶችን የመከከል አደጋን ይቀንሳል፡፡	1. እጅግ በጣም አልስመናም 2. አልስመናም 3. ገለልተኛ 4. እስመናላሁ 5. በጣም እስመናላሁ
6	የቆሰሉ ጥቶች እና እጆች ያሏቸው ትኩስ ምርቶች ሻጮች ፍራፍሬዎችን እና አትክልቶችን መክገፍ ማቆላቆል ለትክክል ከሽፈት ብቻ ነው፡፡	1. እጅግ በጣም አልስመናም 2. አልስመናም 3. ገለልተኛ 4. እስመናላሁ 5. በጣም እስመናላሁ
7	ደህንነት የተጠበቀ ምግብ (ፍራፍሬዎችና አትክልቶች) አያያዝ ለሥራዬ አስፈላጊ የሆኑ ምርቶች አካል ነው፡፡	1. እጅግ በጣም አልስመናም 2. አልስመናም 3. ገለልተኛ 4. እስመናላሁ 5. በጣም እስመናላሁ
8	የጤና ትምህርት የአንጀት ጥገና እና የባክቴሪያ ኢንፎክሽኖች ስርጭትን ሊቀንስ ይችላል፡፡	1. እጅግ በጣም አልስመናም 2. አልስመናም 3. ገለልተኛ 4. እስመናላሁ 5. በጣም እስመናላሁ
9	ንፅህና ከሌላቸው አሠራሮች በኋላ ወዲያው እጅን መታጠብ አስፈላጊ ነው፡፡	1. እጅግ በጣም አልስመናም 2. አልስመናም 3. ገለልተኛ 4. እስመናላሁ 5. በጣም እስመናላሁ
10	እጅን በመታጠብ ጊዜ ሳሙን መጠቀም የአንጀት ጥገና ተሐዋስያን እና የባክቴሪያ ኢንፎክሽኖች መከላከል	1. እጅግ በጣም አልስመናም 2. አልስመናም 3. ገለልተኛ 4. እስመናላሁ 5. በጣም

ይችላል፡፡	እስማላሁ
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መፍራፍሬ እና የአትክልቶች ሽጮች ፍራፍሬ እና አትክልቶች ደህንነት ስልጠናዎች ግምገማ

ተ.ቁ	ጥያቄ	ምላሾች
1.	ፍራፍሬዎችን እና አትክልቶችን ከመያዝ በፊት እጅዎን ወዲያው ይታጠባሉ።	1. በጭራሽ 2. አልፎአልፎ 3. አንዳንድ ጊዜ 4. አብዛኛው ጊዜ 5. ሁል ጊዜ
2.	ትክክለኛውን የእጅ መታጠቢያ ሂደቶች ይከተላሉ።	1. በጭራሽ 2. አልፎአልፎ 3. አንዳንድ ጊዜ 4. አብዛኛው ጊዜ 5. ሁል ጊዜ
3.	መጻዳጃ ቤቱን ለመጻዳጃት ይጠቀማሉ።	1. በጭራሽ 2. አልፎአልፎ 3. አንዳንድ ጊዜ 4. አብዛኛው ጊዜ 5. ሁል ጊዜ
4.	ከመጻዳጃ ቤት በኋላ እጅዎን በሰሜን / በአመቺ ይታጠባሉ።	1. በጭራሽ 2. አልፎአልፎ 3. አንዳንድ ጊዜ 4. አብዛኛው ጊዜ 5. ሁል ጊዜ
5.	በመታመን ትወይም እጆችዎ ላይ ቁስሎች ሲኖሩ ፍራፍሬዎችን እና አትክልቶችን ይይዛሉ።	1. በጭራሽ 2. አልፎአልፎ 3. አንዳንድ ጊዜ 4. አብዛኛው ጊዜ 5. ሁል ጊዜ
6.	የጣትዎን ጥፍር በመደበኛነት ትይቆርጡ።	1. በጭራሽ 2. አልፎአልፎ 3. አንዳንድ ጊዜ 4. አብዛኛው ጊዜ 5. ሁል ጊዜ
7.	ፍራፍሬዎችን እና አትክልቶችን በመይዘት/በሚሽጠበት ጊዜ ጉርዎን ማለብ/ከፊያ (ኬፕ) ተሸፍኑ ወይን ወይን	1. በጭራሽ 2. አልፎአልፎ 3. አንዳንድ ጊዜ 4. አብዛኛው ጊዜ 5. ሁል ጊዜ
8.	አገልግሎትዎን ከጨረሱ በኋላ የሥራ ታዎን ያፀዳሉ።	1. በጭራሽ 2. አልፎአልፎ 3. አንዳንድ ጊዜ 4. አብዛኛው ጊዜ 5. ሁል ጊዜ
9.	አፍንጫዎን ከሽሹ ወይም ከሌሎች ጋር ትዎን ከከፍተኛ ጊዜ እጅ ይታጠባሉ።	1. በጭራሽ 2. አልፎአልፎ 3. አንዳንድ ጊዜ 4. አብዛኛው ጊዜ 5. ሁል ጊዜ
10.	ከሽያጭ ፊት ምርቶችን (ፍራፍሬዎችን እና አትክልቶችን) ታጥባለህ/ሽ።	1. በጭራሽ 2. አልፎአልፎ 3. አንዳንድ ጊዜ 4. አብዛኛው ጊዜ 5. ሁል ጊዜ

ሠ / ምልክታዎቹ በአስተያየት ቁጥጥር ዝርዝር በአትክልትና ፍራፍሬ አቅራቢዎች ይከናወናሉ

የታዩ ፍራፍሬዎችና አትክልቶች ደህንነት ትተግባር	በቂ / በቂ ያልሆነ
1. የሽጮች ግልን ፅሁፍ የተጠበቀ እና የተጠራጠረ ቅጣት ምን ያጠቃልላል፡፡ ጥፍሮችን ደህን እና አጭናቸው	1. በቂ 2. በቂ ያልሆነ
2. የቆዳ ቁስሎች ወይም በሽታዎች / የበሽታ ምልክቶች ሽጮች ላሉት ጊዜ፡፡	1. በቂ 2. በቂ ያልሆነ
3. በጉብኝት ወቅት ከአፍንጫ ከዓይን፣ ከጆሮ እና ከሰገላ ማወጣት ስሜት ላይ ለምን፡፡	1. በቂ 2. በቂ ያልሆነ
4. የንደህምርቶች (ፍራፍሬዎች እና አትክልቶች) ንፅህና መያዝ፡፡	1. በቂ 2. በቂ ያልሆነ

5.3. Unka gafanno “KAP” afaanii Afaan oromootin qophaa’e

Gabatee 7.Gaaffannoo sakkata’insaa “KAP” Gurgurtoota Kuduraa fi muduraa Magaala Jimmaatti Gandoota Filataman sadan(Jireen,Booree fi Hora Gibee) irratti Maxantuulee garaa fi Baakteriyaa Faalama Kuduraa fi muduraa Gabaaratti gurguraman fidan qorachuudhaaf kan fayyadamnu

A.Ragaa bu’uuraa haala walii galaa, Jijiramootafi haariiroo Qabxii Beekumsa, Ilaalchaa fi Gocha (KAP) Gurgurtoota Kuduraa fi muduraa ilaalchisee

Lakk	Gaaffii	Deebii
1	Koodii Saamudaa	_____
2	Saala	1.Dhiira 2.Dubar
3	Umurii Deebii kennaa/Hirmaataa/ttu	Waggaa_____
4	Guyyaa Saamudni Munaanname	_____
5	Bakka Qorannoo	_____
6	Maqaa Gabaa	
7	Gosa Gabaa	1. Giroosarii 2. Gabaa Dirree
8	*Gosa muduraa fi kuduraa	
9	Madda muduraa fi kuduraa	1. Qonnaan bultoota 2. Daldaaloota 3. Lafa kuduraa fi muduraa dhunfaa irraa
10	Bakka/Teessoo muduraa fi kuduraa itti gurguramaa jiru	1.Lafa irratti 2. Madardarraa irratti 3. Minjaala irratti 4. Gaarii dhibamee deemu irraa
11	Muduraa fi kuduraa gurguramaa jiru miicameetii?	2. Eyyeni 2. Lakki
12	Bishaan akkamii mana keessatti fayyadamaa jirtu (muduraa fi kuduraa micuu fi k.k.f tuuf)	1. Kan Ujummoo 2. Kan boollaa 3. Kan Lagaa

13	Haala Gaa'ila ilaalchisee	1. Hin heerumne /hin fuune 2.Gaa'ila kan heekte/hiike 3. Kan heerumte/fuudhe 4. Abbaan mana /haatii manaa kan du'e
14	Sadarkaa barumsaa gurgurtoota	1.sagantaan alaa 2. Sad.tokkoffan baradhe. 3. Sad. Lamaffan baradhe 4.Kolleji/yuniiversitii
15	Galii Ji'aa argatan (Akka Birri Itoophiyaatti)	_____
16	Leenjii Nageenya muduraa fi Kuduraa fudhachuu	1.Lakki 2.Eeyyeni
17	Muuxannoo Hojii(Waggaadhaan)	1. < 1 3. 3-5 2. 1-2 4. > 5

B. Gaaffannoo waa'ee Nageenya muduraa fi Kuduraa Sakkata'iinsa Beekumsa Gurgurtoota muduraa fi kuduraa ilaalchisee

1.	Hojii dura harka dhiqachuun balaa faalama Kuduraa fi muduraa ni xiqqeessa	1.Dhugaa 2.Soba 3.Hin beeku/hin yaadadhu
2.	Golga harkaa godhachuun qulqullina harka keenyyaf bakka bu'iinsa hin qabu	1.Dhugaa 2.Soba 3.Hin beeku/hin yaadadhu
3.	Nageenya dhabuunGurgurtoota Kuduraa fi Muduraa faalama Kuduraa fi muduraa Maxxantootaan dhufaniin sababa ta'uun dhukkubaf nama saaxiluu danda'a.	1.Dhugaa 2.Soba 3.Hin beeku/hin yaadadhu
4.	Sadarkaa Nageenya Gurgurtoota Kuduraa fi Muduraa Yeroo yeroon sakkata'un barbaachisaadha.	1.Dhugaa 2.Soba 3.Hin beeku/hin yaadadhu
5.	Kuduraa fi Muduraa bishaan lagaan dhiquu fiQuncisuun qulqullina kuduraa fi muduraa nyaachuuf gahaadha.	1.Dhugaa 2.Soba 3.Hin beeku/hin yaadadhu

6	Kuduraa fi muduraa sirritti bilchaate faaltoota/maxantoota nyaata faaluu dhukkuba fiduu danda'an irraa gutuman guttutti bilisadha.	1.Dhugaa 2.Soba 3.Hin beeku/hin yaadadhu
7	Gurgurtoota Kuduraa fi Muduraa harki isaanii murame ykn madaa qabu tuttuqii fi qabannaa kuduraa fi muduraa irraa dhorkuun barbaachisaa miti.	1.Dhugaa 2.Soba 3.Hin beeku/hin yaadadhu
8	Kuduraa fi Muduraa dheedhii nyaachuun sababa dhukkubaa ta'uu danda'a.	1.Dhugaa 2.Soba 3.Hin beeku/hin yaadadhu
9	Qulqullina dhabuun Kuduraa fi Muduraa sababa dhukkuba raammolee maxantota garaa fi baacteeriyaa ta'uu danda'a.	1.Dhugaa 2.Soba 3.Hin beeku/hin yaadadhu
10	Yeroo dhukkuba gogaa qaamaa (madaa qaamaa) qabaman hanga fayyamutti bakka hojii irraa fagaachuun barbaachisaa dha.	1.Dhugaa 2.Soba 3.Hin beeku/hin yaadadhu
11	Balfa karaa seeraan alaa naannoo bakka hojiitti gatamu Sababa orgaanizimoota kuduraa fi muduraa faaluu danda'an ta'uu danda'a.	1.Dhugaa 2.Soba 3.Hin beeku/hin yaadadhu
12	kuduraa fi muduraa dheedhii nyaachuun faayidaa qofa osoo hin ta'in miidhaas qaba.	1.Dhugaa 2.Soba 3.Hin beeku/hin yaadadhu

C.Gaaffannoo waa'ee Nageenya muduraa fi Kuduraa Sakkata'iinsa Ilaalcha gurgurtoota muduraa fi kuduraa ilaalchisee

	Gaaffii	Deebii
1	Irra deddebiin harka dhiqachuun yeroo hojii balleessa	1. Sirritin irratti walii gala 2.waliin gala 3. Yaada hin qabu4. walii hin galu 5.Sirritin irratti walii hin galu
2	Karaa sirrii hin taaneen kuduraa fi muduraa kuusuun balaa fayyaa ta'uu danda'a.	1. Sirritin irratti walii gala 2. Waliin gala 3. Yaada hin qabu 4. Walii hin galu 5. Sirritin irratti walii hin galu

3	Eggannoon qulqullina bakka hojii balaa faalama kuduraa fi muduraa hir'isuu danda'a.	1. Sirrirtin irratti walii gala 2. Waliin gala 3. Yaada hin qabu 4. Walii hin galu 5. Sirrirtin irratti walii hin galu
4	Idileen duraa duuba hojii to'anoo faalama nyaataa (kuduraa fi muduraa) sirritti hojii irra olchuun dawoo gahaa dhukkuba irraa bararefamuu fayyadamtootaa ti	1. Sirrirtin irratti walii gala 2. Waliin gala 3. Yaada hin qabu 4. Walii hin galu 5. Sirrirtin irratti walii hin galu
5	Osoo kuduraa fi muduraa hin tuqin dura harka dhiqachuun balaa faalama kuduraa fi muduraa hir'isuu danda'a.	1. Sirrirtin irratti walii gala 2. Waliin gala 3. Yaada hin qabu 4. Walii hin galu 5. Sirrirtin irratti walii hin galu
6	Gurgurtootni kuduraa fi muduraa kanneen madaa quba fi harka irraa qaba kuduraa fi muduraa kan tuttuqaa qaban yoo madaa isaanii sirriitti haguugan qofa dha.	1. Sirrirtin irratti walii gala 2. Waliin gala 3. Yaada hin qabu 4. Walii hin galu 5. Sirrirtin irratti walii hin galu
7	Eggannoon kuduraa fi muduraa qabuun qaama barbaachisaa gahee hojii gurgutootaa fi dirqama gurgurtootaa ti.	1. Sirrirtin irratti walii gala 2. Waliin gala 3. Yaada hin qabu 4. Walii hin galu 5. Sirrirtin irratti walii hin galu
8	Barumsa fayyaa argachuun tattamsa'ina dhukkuba raammolee garaa fi baakteeriyaa dhufan hir'isuu danda'a.	1. Sirrirtin irratti walii gala 2. Waliin gala 3. Yaada hin qabu 4. Walii hin galu 5. Sirrirtin irratti walii hin galu
9	Erga hojii adda addaa hojenneen booda harka keenya dhiqachuun barbaachisaa dha.	1. Sirrirtin irratti walii gala 2. Waliin gala 3. Yaada hin qabu 4. Walii hin galu 5. Sirrirtin irratti walii hin galu
10	Yeroo harka keenya dhiqannuu saamunaa fayyadamuun dhukkuboota raammolee garaa fi baakteeriyaa dhufan nurraa ittisuu danda'a.	1. Sirrirtin irratti walii gala 2. Waliin gala 3. Yaada hin qabu 4. Walii hin galu 5. Sirrirtin irratti walii hin galu

D.Gaaffannoo waa'ee Nageenya muduraa fi Kuduraa Sakkata'iinsa Gocha gurgurtoota muduraa fi kuduraa ilaalchisee

Gaaffii	Deebii
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1.	Attaatamaan osoo kuduraa fi muduraa hin tuqin /hin qabin harka kee ni dhiqattaa?	1.Lakki 2.Baay'ee darbe darbee 3.Yeroo tokko tokko 4.Yeroo baay'ee 5.Yeroo hunda
2.	Yeroo harka dhiqannaa duraa duuba sirrii harka dhuqannaa ni hordoftaa?	1.Lakki 2.Baay'ee darbe darbee 3.Yeroo tokko tokko 4.Yeroo baay'ee 5.Yeroo hunda
3	Mana fincaanii ni fayyadamtaa?	1.Lakki 2.Baay'ee darbe darbee 3.Yeroo tokko tokko 4.Yeroo baay'ee 5.Yeroo hunda
4.	Mana fincaanii erga fayyadamteen booda harka kee saamunaa/daaraan ni dhiqattaa?	1.Lakki 2.Baay'ee darbe darbee 3.Yeroo tokko tokko 4.Yeroo baay'ee 5.Yeroo hunda
5.	Yeroo si dhukkubuu ykn yeroo qubni/harki ke murame kuduraa fi muduraa ni qaqqabattaa/ni tuttuqxaa?	1.Lakki 2.Baay'ee darbe darbee 3.Yeroo tokko tokko 4.Yeroo baay'ee 5.Yeroo hunda
6.	Yeroo yeroo qeensa harka kee ni qorattaa?	1.Lakki 2.Baay'ee darbe darbee 3.Yeroo tokko tokko 4.Yeroo baay'ee 5.Yeroo hunda
7	Rifeensa mataa kee guutumaa tuututti ni haguugdaa yeroo kuduraa fi muduraa tuttuqxu/gurgurtu?	1.Lakki 2.Baay'ee darbe darbee 3.Yeroo tokko tokko 4.Yeroo baay'ee 5.Yeroo hunda
8	Bakka hojii kee ni qulqullesitaa erga hojii xummurtee booda?	1.Lakki 2.Baay'ee darbe darbee 3.Yeroo tokko tokko 4.Yeroo baay'ee 5.Yeroo hunda
9	Harka kee ni dhiqatta erga funyaan kee qulqullesitee ykn erga qaama kee hooqxee booda?	1.Lakki 2.Baay'ee darbe darbee 3.Yeroo tokko tokko 4.Yeroo baay'ee 5.Yeroo hunda
10	kuduraa fi muduraa osoo hin gurgurin duran ni miicitaa?	1.Lakki 2.Baay'ee darbe darbee 3.Yeroo tokko tokko 4.Yeroo baay'ee 5.Yeroo hunda

ANNEX-IV

4. LABORATORY PROCEDURES

4.1. Sedimentation Procedure

A portion (200 g) of each vegetable and fruit will be washed separately in 500 mL of normal saline

1. Keep overnight sedimentation of the washing solution,
2. Sieve the washing solution to remove undesirable matters
3. Then 15 mL of the sediment will then be transferred to a centrifuge tube
4. The tube will be centrifuged at 3000 rpm for five minutes.
5. After centrifugation, the supernatant will be decanted carefully without shaking.
6. Then the sediment will be agitated gently by hand for redistributing the parasitic stages.
7. Finally, the sediment will be examined under a light microscope using $\times 10$ and $\times 40$ objectives.
8. Report the finding using the prepared form

4.2. Modified Zeihl-Neelsen staining technique procedure

1. Prepare a smear with 1 to 2 drops of segmented solution on the slide and dry on slides. Do not make the smears too thick.
2. Fix with absolute methanol for 30 seconds.
3. Stain with carbon fuschin for one minute. Rinse briefly with distilled water and drain.
4. Destain with acid alcohol for two minutes. Rinse with distilled water and drain.
5. Counterstain with metylen blue for 2 minutes and rinse briefly with distilled water and drain.
6. Dry on a slide for 5 minutes. Mount with a coverslip using desired mounting media.
7. Examined 200 to 300 fields using 40x or higher objectives. To confirm internal morphology, use 100x oil immersion objective.

4.3. Cultures' Procedure

1. About 25g of vegetable and fruit samples were rinsed thoroughly with 225 ml sterile distilled water/sterile 0.1% (W/V) bacteriological peptone water (oxid) for 3 minutes
2. Tenfold serial dilution of 10^{-1} , 10^{-2} , 10^{-3} , 10^{-4} and 10^{-5} was made
3. 0.1 ml of the suspension from each dilution was pipetted into MacConkey agar, Salmonella - Shigella agar, and Mannitol Salt agar using the streak Plate Technique.
4. The plates were allowed to solidify, inverted and incubated at 37 °C for 24 h for colony formation.
5. Distinctive morphological properties of each pure culture such as colony form, elevation of colony and colony margin were observed. Bacterial pathogens were isolated and identified on the basis of morphological, cultural and different biochemical tests

4.4. Principle of MacConkey Agar

MacConkey Agar is used for the isolation of gram negative enteric bacteria and differentiation of lactose fermenting from lactose non fermenting gram negative bacteria. Pancreatic digest gelatin and peptone (meat and casein) provide the essential nutrients. Vitamins and nitrogenous factors require for growth of microorganisms. Lactose monohydrate is the fermentable source of carbohydrate. The selective action of this medium is attributed to crystal violet and bile salts, which are inhibitory to most species of gram positive bacteria. Sodium chloride maintains the osmotic balance in the medium. Neutral red is a PH indicator that turns red at a PH below 6.8 and is colorless at any PH greater than 6.8. Agar is the solidifying agent.

4.5. Principle of Salmonella Shigela Agar

Salmonella Shigela Agar comprises of bile salts, sodium citrate, and brilliant green, enzymatic digest of casein, beef extract, enzymatic digest of animal tissue, thiosphate, ferric citrate, neutral red and agar. The inclusion of bile salts, sodium citrate, brilliant green serve to inhibit gram positive, Coliform organisms and inhibit swarming proteus Spp, while allowing salmonella spp to grow. Enzymatic digest of casein, beef extract, enzymatic digest of animal tissue provide source of nitrogen, carbon, and vitamins required for organisms growth. Lactose serve as carbohydrate source in salmonella shigela agar. Differentiation of enteric organisms is achieved by the incorporation of lactose in the medium. Organisms which ferment lactose produce acid which, in the presence of the neutral red indicator, results in the formation of red/pink colonies. Lactose non fermenters form colorless colonies. **Thiosphate** and **ferric citrate** permit detection of hydrogen sulfide by the production of colonies with black centers. **Neutral red** turns red in the presence of an acid PH, thus showing fermentation has occurred.

ANNEX-Five

6.1. Assurance of principal investigator and Declaration

The undersigned agrees to accept responsibility for the scientific ethical and technical conduct of the research project and for provision of required progress reports as per terms and conditions of the Faculty of Health Sciences in effect at the time of grant was forwarded as the result of this application.

Declaration Sheet

This thesis is my original work, has not been presented for a degree or a master in any University.

This thesis entitled “INTESTINAL PARASITIC AND BACTERIAL CONTAMINATION OF FRESH/RAW VEGETABLES AND FRUITS SOLD IN OPEN-AIRED MARKETS IN SELECTED PERI-URBANKEBELES OF JIMMA TOWN, SOUTHWEST ETHIOPIA.” had not been presented in Institute of Health, Jimma University from the study area pre-urban kebeles of Jimma town.

Submitted by **Signature** **Date**
Jiru Batu _____ ___/___/___

Researcher

Major Advisor **Signature** **Date**
DR. AHMED Z. (PhD, ASSOCIATE PROFESSOR) 1, _____ ___/___/___

Co-Advisor
MR. TARIKU B. (BSC, MSC) 2. _____ ___/___/___

DR. TESHOME D. (Ph.D, ASSISTANT PROFESSOR) 3 _____ ___/___/___

Internal examiner _____

Signature _____ Date _____

External examiner _____

Signature _____ Date _____

ANNEX-Five

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Name of the pricipal Investigator	Signature	Date
Jiru Batu	_____	___/___/___

Approval of the first Advisor	Signature	Date
DR. AHMED Z. (PhD, ASSOCIATE PROFESSOR) 1,	_____	___/___/___

Approval of co- advisors	Signature	Date
MR. TARIKU B. (BSC, MSC) 2.	_____	___/___/___
DR. TESHOME D. (Ph.D, ASSISTANT PROFESSOR) 3	_____	___/___/___

Approval by Assessor	Signature	Date
_____	_____	___/___/___

Head of Department	Signature	Date
_____	_____	___/___/___