**HARAMAYA UNIVERSITY**

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# Magnitude, Associated Factors and Antimicrobial Susceptibility Patterns of *Salmonella* and *Shigella* Species among Human Immunodeficiency Virus-Infected Patients Attending Treatment at Hiwot Fana Comprehensive Specialized University Hospital, Harar, Eastern Ethiopia

**MSc Thesis**

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**Harar, Ethiopia**

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# Magnitude, Associated Factors and Antimicrobial Susceptibility Patterns of *Salmonella* and *Shigella* Species among Human Immunodeficiency Virus-Infected Patients Attending Treatment at Hiwot Fana Comprehensive Specialized University Hospital, Harar, Eastern Ethiopia

**A Thesis Submitted to the School of Medical Laboratory Sciences, College of Health and Medical Sciences, Haramaya University**

**For Partial Fulfillment in Requirements for the Degree of Master of Science in Medical Microbiology**

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**Co-adviser: Dr. Desalegn Admassu (Ph.D., Associate professor)**

**April 2025**

**Harar, Ethiopia**

# APPROVAL SHEET

## POST GRADUATE PROGRAMS DIRECTORATE

I certify that I have read and evaluated this thesis, entitled Magnitude, Associated Factors and Antimicrobial Susceptibility Patterns of *Salmonella* and *Shigella* Species among Human Immunodeficiency Virus-Infected Patients Attending Treatment at Hiwot Fana Comprehensive Specialized University Hospital, Harar, and Eastern Ethiopia, under my guidance by Sisay Geremew Gurmu. I recommend that it be submitted as fulfilling the thesis requirement.

Mr. Zelalem Teklemariam (MSc, Associate professor) ---------- ----------

Major advisor: Signature Date

Dr. Desalegn Admassu (Ph.D., Associate professor) ---------- ----------

Co-adviser: Signature Date

As a member of the board of examiners of the Medical Microbiology Thesis Open Defense Examination, I certify that I have read and evaluated the thesis prepared by Sisay Geremew Gurmu and examined the candidate. I recommend that the thesis be accepted as fulfilling the thesis requirements for the Master of Medical Microbiology degree.

Chair Person: _____ Signature: _____ Date: _____

Internal examiner: _____ Signature: _____ Date: _____

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Final approval and acceptance of the thesis are contingent upon the submission of its final copy to the Council of Graduate Studies (CGS) through the School of Graduate Committee (SGC).

# DEDICATION

I dedicate this thesis work to the almighty God my creator, my strong pillar, my source of inspiration, wisdom, knowledge, and understanding.

## STATEMENT OF THE AUTHOR

By my signature below, I declare and affirm that this thesis is my work. I have followed all ethical principles of scholarship in the preparation, data collection, data analysis, and completion of this thesis. All scholarly matter that is included in the thesis has been given recognition through citation. I affirm that I have cited and referenced all sources in this document. Every serious effort has been made to avoid any plagiarism in the preparation of this thesis. This thesis is submitted in partial fulfillment of the requirement for a master's degree from the School of Graduate Studies at Haramaya University. The thesis is deposited in the Haramaya University Library and is made available to borrowers under the rules of the library. I solemnly declare that this thesis has not been submitted to any other institution anywhere for the award of any academic degree, diploma, or certificate. Brief quotations from this thesis may be used without special permission provided that accurate and complete acknowledgement of the source is made. Requests for permission for extended quotations from, or reproduction of, this thesis in whole or in part may be granted by the Head of the School or Department or the Dean of the School of Graduate Studies when in his or her judgment the proposed use of the material is in the interest of scholarship. In all other instances, however, permission must be obtained from the author of the thesis.

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# BIOGRAPHICAL SKETCH

I was born in 1991G.C in Lega Dadi town. I have completed my elementary school at Dale Dambel Elementary School. I have attended my secondary school in Sendafa secondary preparatory school. After completion of my preparatory school, I joined Wollega University. At Wollega University, I have studied Medical Laboratory Sciences. I got my first BSc degree in Medical Laboratory Science in June 2014 G.C. My cumulative GPA was 3.67. I have been working as a laboratory technologist from 2014 to 2022 G.C. My master grade report was 3.71.

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First, I would like to thank Haramaya University, School of Medical Laboratory Science, my advisor Mr. Zelalem Teklemariam, and co-advisers Dr. Desalegn Admassu and Mr. Fikru Tebeje for their great support, guidance, and valuable contribution to the development of this research defense, and all staff of the Medical Microbiology department for contributing materials or idea for research defense development. Lastly, I would like to acknowledge my family, friends, study participants, data collectors, and Hiwot Fana Comprehensive Specialized Hospital workers for their great help and encouragement. Finally, I would like to thank all who have directly or indirectly contributed to this research defense.

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# LIST OF ACRONYMS AND ABBREVIATIONS

| | |
|---|---|
| AIDS | Acquired Immune- Deficiency Syndromes |
| ART | Anti-Retroviral Therapy |
| ATCC | American Type Culture Collector |
| CDC | Center for Disease Prevention Control |
| CLSI | Clinical and Laboratory Standards Institute |
| HFCSUH | Hiwot Fana Comprehensive Specialized University Hospital |
| HIV | Human Immunodeficiency Virus |
| MDR | Multi-Drug Resistance |
| UK | United Kingdom of England |
| XLD | Xylose Lysine Deoxycholate |

# ABSTRACT

**Background:** *Salmonella* and *Shigella* species are the main causative agents of diarrhea in people living with Human Immunodeficiency Virus. The magnitude, associated factors, and antibiotic sensitivity tests of *Salmonella* and *Shigella* species were not studied on Human Immunodeficiency Virus infected patients in the study area.

**Objective:** To determine the magnitude, associated factors, and antimicrobial susceptibility patterns of *Salmonella* and *Shigell*a species among Human Immunodeficiency Virus-infected patients attending treatment at Hiwot Fana Comprehensive Specialized University Hospital, Harar, Ethiopia from February 01 to April 30, 2024.

**Methods:** An institutional-based cross-sectional study was conducted on 235 Human Immunodeficiency Virus-infected Patients with complaints of diarrhea. Data were collected through a face-to-face interview using a structured questionnaire. Stool samples were collected, inoculated to selenite F broth, and subcultured to MacConkey and Xylose Lysine Deoxycholate agar, and identified using biochemical tests. The modified Kirby-Bauer disk diffusion techniques were used to determine drug susceptibility patterns. Data were entered into Epi data version 3.1, and analyzed using Statistical Package for Social Sciences version 26. Logistic regression was performed to check an association between variables. A P-value < 0.05 was considered statistically significant.

**Results:** The overall prevalence of *Salmonella* and *Shigella* species was 7.7 %( 95%CI 4.0-11.0). *Salmonella* species 4.7 %( 95% CI: 2.0-7.0) and *Shigella* species 3 %( 95% CI: 1.0-5.0). The highest resistance was seen for *Salmonella* species against Ampicillin (100%), Nalidixic acid (54.5%), Cotrimoxazole (45.5%) and Chloramphenicol (45.5%), for *Shigella* species against Ampicillin (100%), Tetracycline(85.7%), and Nalidixic acid (71.4%). and Co-trimoxazole (57.1%). unprotected source of water (AOR = 8.10, 95% CI: 1.54-42.61, p = 0.013), and clinical stage3 and4 (AOR = 27.27, 95% CI: 27.27(14.13-40.41), p = 0.001) were factors associated with *Salmonella* and *Shigella* infection .

**Conclusion:** High prevalence of *Salmonella* and *Shigella* species were found. There were higher drug resistance and multidrug-resistant patterns. Providing safe potable water and improving clinical status are recommended.



**Keywords:** *Salmonella, Shigella,* Drug resistance, Human Immunodeficiency Virus, Ethiopia

# 1. INTRODUCTION

## 1.1. Background

Human Immunodeficiency Virus (HIV) continues to be a major global public health issue. Worldwide, an estimated 38.4 million people are living with HIV of which 36.7 million are adults (15 years or older) and 1.7 million are children (0–14 years). About 28.7 million were receiving Anti-Retroviral Treatment (ART) in 2021, 25.6 million were in Africa, 650, 000 people died from HIV-related causes and 1.5 million people acquired new HIV in 2021 (WHO HIV REPORT, 2022).

The most prevalent bacterial pathogens in HIV patients are *E.coli, Shigella*, *Campylobacter*, and *Salmonella* species (especially enteric serotypes). Campylobacter infections alone occurrence is 40 times higher among AIDS patients than among non-infected individuals (Geissler *et al.*, 2017).

Factors associated with *Salmonella* among HIV-positive individuals are low CD4 count (*Udoh et al.*, 2023). HIV binds to the CD4 molecule on the surface of helper T-cells and replicates within them. This results in the destruction of CD4+ T-cells and leads to a steady decline in this population of T-cells (Vidya Vijayan *et al.*, 2017). This decline in CD4+ T-cell count puts HIV patients at increased risk of being infected with *Salmonella* and *Shigella* infection ((Ngalani *et al*., 2019); (Udoh *et al.*, 2023);(Sahoo *et al.*, 2018))

*Salmonella* (*Salmonella enterica serotypes Typhimurium and Enteritidis), and Shigella* species are the common causes of diarrhea among HIV-infected patients. HIV patients are at increased risk for developing salmonellosis and they have the incidence rates of salmonellosis as 20–100 folders higher than the non-HIV patients. *Shigella* is more common among HIV patients and might occur in both mild and severe cases of clinical shigellosis*(*Rameshkumar and Arunagirinathan, 2018*)*. Disseminated infections caused by *Salmonella* are higher in HIV-infected populations(Koehler and Chaisson, 2020).

*Salmonella* Typhi and *Salmonella* Paratyphi cause life-threatening typhoid fever and paratyphoid fever, respectively, with estimated annual mortalities of over 150,000 worldwide(Sang *et al.,* 2024).

Globally in 2017, non-typhoidal *salmonella* caused 77,500 deaths of that 18,400 death were attributable to HIV. In high-income countries, death among people with HIV was 0·68 per thousand. In sub-Saharan Africa, the cause of death was 17·08 per thousand (Stanaway *et al.,* 2019).

*Salmonella* and *Shigella* can be diagnosed by stool culture on selective media such as Xylose Lysine Deoxycholate agar, Salmonella-Shigella agar, and Hektoen agar(Parija, 2023). Molecular tests such as single specific primer-loop mediated isothermal amplification (SSP-LAMP)(Mollasalehi *et al.,* 2022), multiplex fluorescent polymerase Chain Reaction (PCR) assay (He *et al.,* 2022), and High-resolution melting (HRM) assay are used for diagnosis of *Shigella species* (Pakbin *et al.,* 2022).

The initial treatment of choice for *Salmonella* infection for people living with HIV is Ciprofloxacin 500–750 mg PO (or 400 mg IV) every 12 hours. Alternatives Trimethoprim 160 mg/sulfamethoxazole 800 mg (PO or IV) every 12 hours, Ceftriaxone IV 1 g every 24 hours, or Cefotaxime IV 1 g every 8 hours. For shigellosis Ciprofloxacin 500–750 mg PO for 7 to 10 days or 400 mg IV every 12 hours. Alternative Trimethoprim 160 mg/sulfamethoxazole 800 mg PO or IV every 12 hours((of the Infectious *et al.,* 2022*)*;(AIDSinfo, 2019))

Typhoid fever is caused by *Salmonella typhi. Non-typhoidal* salmonellosis (NTS) refers to any illnesses caused to humans by all serotypes of *Salmonella*, except for the distinct typhoidal serotypes *S.typhi* and *S. Paratyphi A-C*. Invasive Non-Typhoidal *Salmonella* (iNTS) are *S. typhimurium* and *S. enteritidis*; however, other serotypes such as Choleraesuis and Dublin are also known to cause invasive diseases in humans (Kurtz *et al.,* 2017).

In sub-Saharan Africa Since 2001, multidrug resistance has been observed in 75% of Nontyphoidal *salmonella* (NTS) isolates. Third-generation cephalosporin resistance emerged in all sub-Saharan

African regions and was present in 5% after 2010. Fluoroquinolone non-susceptibility emerged in all sub-Saharan African regions but did not increase over time (Tack *et al.*, 2020). In Ethiopia, different studies are reporting resistance to commonly used antibiotics like Ampicillin, Tetracycline, Chloramphenicol, and Sulfamethoxazole– Trimethoprim. One study conducted at Arba Minch shows that Multidrug resistance was detected in *Shigella* species (100%). *Shigella* species show resistance to Ampicillin (100%), Gentamicin (100%), Erythromycin (100%), and Ceftazidime (100%). In addition, (50%) resistance to four anti-biotic Tetracycline (50%), Ciprofloxacin (50%), Co-trimoxazole (50%), and Ceftriaxone (50%)(Ayele *et al.*, 2020). Another cross-sectional study conducted at Dessie on 112 HIV infected from January to March 2018 shows *Salmonella* species were resistant to Amoxicillin (100%), Ampicillin (100%), Tetracycline (47%), Co-trimoxazole (41%), Chloramphenicol (35.2%), and Nalidixic acid (17.6%) (Belay *et al.*, 2020). Antibiotic resistance surveillance is important to monitor drug resistance in different parts of the world. This study will try to determine the magnitude, associated factors, and antimicrobial susceptibility patterns of *Salmonella* and *Shigella* species among HIV patients attending treatment at Hiwot Fana Comprehensive Specialized University Hospital, Harar, Ethiopia.

## 1.2 . Statement of Problem

Human Immunodeficiency Virus (HIV)-infected individuals are highly vulnerable to various opportunistic infections. Multi-drug-resistant (MDR) pathogens are relentlessly multiplying in HIV patients and thus become an important circulating source of infection in the community (Rameshkumar and Arunagirinathan, 2018). HIV-infected patients have an increased risk of bacteria (by 20- to 100-fold) and mortality (by up to 7-fold) compared with HIV-negative individuals(Masur, 2017).

Globally in 2017, death due to non-typhoidal *Salmonella* invasive disease, among people living with HIV was 18·40 per thousand, with a mortality rate of 2.31 per million. The proportion of cases attributable to HIV was 24·3% (Stanaway *et al.*, 2019). In 46 out of 53 European regions that belong to Europe in 2020, an estimated 104,765 are living with HIV from that *Salmonellosis* cases reported was 52,702, with Hospitalizations of 6149, and deaths 57(Russotto *et al.*, 2022). Study conducted at England from 2004 to 2015,shows out of 88 664 adults age >15 living with HIV , 16,244 *Shigella* cases were reported(Mohan *et al.*, 2018).

In high-income countries, non-typhoidal *salmonella* death among people with HIV was 0·68 per thousand, with a mortality rate of 0·41 per million, and the proportion of cases attributable to HIV was 50·0%. In Latin America and the Caribbean, death per thousand was 0·23, mortality per million was 0·38, and the proportion of cases attributable to HIV was 22·3% (Stanaway *et al.*, 2019).

In sub-Saharan Africa in 2017, in people with HIV infection due to non-typhoidal *salmonella* invasive disease deaths were 17·08 per thousand, and mortality rates of 23·66 per million, and Proportion of cases attributable to HIV was 26·2%. In North Africa and the Middle East, death per thousand was 0·03, mortality per million was 0·05, and the proportion of cases attributable to HIV was 1·7% (Stanaway *et al.*, 2019).

In Ethiopia, the prevalence of *Salmonella* species, at Arba Minch was 2.1% (Ayele *et al.*, 2020), at Hawassa was 5.1% (*Kebede et al.,* 2017), at Dessie 5.4% (Belay *et al.*, 2020). The prevalence of *Shigella* species at Addis Ababa was 6.8%(Tadesse *et al.,* 2020), and at Arba Minch was 1.1% (Ayele *et al.*, 2020). Factors associated with *Salmonella* among HIV-positive individuals are CD4 T-cell counts<200 cells/μL (Ayele *et al.,* 2020). The educational status of illiterates, not utilizing latrines, and drinking river or spring water are among the factors associated with *Salmonella* and *Shigella* among HIV (Hlashwayo *et al.,* 2023).

Studies in Ethiopia reveal growing resistance of *Salmonella* and *Shigella* species to commonly used antibiotics. In Hawassa, *Salmonella* isolates showed high resistance to Trimethoprim-Sulfamethoxazole and Chloramphenicol (72% each) (Kebede *et al.,* 2017). In Dessie, *Salmonella* strains were resistant to Amoxicillin and Ampicillin (100%), with varying resistance to other antibiotics, including Tetracycline (47%) and Nalidixic acid (17.6%) (Belay *et al.,* 2020). *Shigella* species demonstrated multidrug resistance, with resistance rates as high as 100% for Ampicillin, Gentamicin, Erythromycin, and Ceftazidime in Arba Minch, and significant resistance to Ciprofloxacin and Ceftriaxone (50% each) (Ayele *et al.,* 2020). A Dessie study highlighted that Ampicillin, Amoxicillin, and Trimethoprim-Sulfamethoxazole are ineffective for treating *Salmonella* and *Shigella*, recommending Ciprofloxacin and Ceftriaxone (Belay *et al.,* 2020). However, limitations like small sample sizes in earlier studies (e.g., in Hawassa) may weaken their conclusions. To address these gaps, the current study increased the sample size to improve reliability and associations.

### 1.3. Significances of the Study

The finding of this study helps to treat HIV patients based on their drug susceptibility pattern when patients are infected with *Salmonella* and *Shigella* species. This study also helps the Harari Regional Health Office, and Organizations working on HIV to get information on drug resistance patterns of *Salmonella* and *Shigella* species to plan for intervention. This study findings will also be used as a baseline information for researchers on these topics to access the precise magnitudes of the problems and implements a solution that alleviates the problems.

## 1.4. Objectives

### 1.4.1. General objective

To determine the magnitude, associated factors, and antimicrobial susceptibility profile of *Salmonella* and *Shigella* species among HIV patients attending treatment at Hiwot Fana Comprehensive Specialized University Hospital, Harar, Ethiopia from February 01 to April 30, 2024.

### 1.4.2. Specific objectives

- To determine the magnitude of *Salmonella* and *Shigella* species among HIV patients
- To determine antimicrobial susceptibility patterns of isolated *Salmonella and Shigella* species among HIV patients
- To identify factors associated with *Salmonella* and *Shigella* species infection among HIV patients

# 2. LITERATURE REVIEW

## 2.1. Magnitude of *Salmonella* species in people living with HIV

A cross-sectional study conducted at an Iraq Hospital on 70 HIV patients from 2020 to 2021 by culture method shows the prevalence of *Salmonella* species was 12.8%. Participants aged 15 to 55 years (Othman *et al.,* 2022).

A prospective study conducted at a tertiary care Hospital in northern India between January 2014 and December 2015 on 300 HIV seropositive cases with diarrhea (study group) and 600 HIV-negative diarrhea cases (control group) shows the prevalence of *Salmonella* species 9(3%) among HIV. Out of the three hundred HIV, seropositive subjects recruited 206 (68.67%) were males and 94 (31.33%) were females. The mean age of the participants in our study was 30.64 ± 13.47 years, and the age ranged between 1 and 65 years(Goel *et al.,* 2018).

In Southeast Asia, East Asia, and Oceania, non-typhoidal *Salmonella* cause deaths among people with HIV per thousand was 0·16, and the mortality rate per million was 0·06. The proportion of cases attributable to HIV was 9·2%. In Central Europe, Eastern Europe, and Central Asia, deaths per thousand were 0·06, mortality rates per million was 0·13, Proportion of cases attributable to HIV was 14·4%, In South Asia death per thousand was 0·17, mortality per million was 0·10, the proportion of cases attributable to HIV was 3·6%. In people with HIV infection, male death per thousand was 9·00, mortality per million was 2·27, proportion of cases attributable to HIV was 23·0%. Female death per thousand was 9·40, mortality per thousand was 2·36, and the proportion of cases attributable to HIV was 25·6% (Stanaway *et al.,* 2019).

An observational study conducted in Mozambique from 1, April 2016, to 28, February 2019, by culture method on 808 HIV patients shows the prevalence of *Salmonella* species was 11%. Male was 57%. The age range of 0–12 months (Kenga *et al.,* 2021).

A cross-sectional study conducted at Cameron from September 2016 to June 2017 on 104 HIV patients(64 females and 40 males) and 48 HIV negative by culture method shows the prevalence

of *Salmonella* species among HIV-positive was 2.88%, compared to HIV-negative which is 4.17%. Their age ranged from 1 to 85 years with mean 36.5 ± 16.6 years (Mabeku *et al.,* 2020).

A cross-sectional study conducted at Nigeria Hospital on 39 HIV patients by culture method shows the prevalence of non-typhoid *Salmonella* was 15%. Females were 72%. Age ranges from 18 to 57 years (Sahal *et al.,* 2020).

A Hospital-based prospective study conducted from April to July 2015 in Nigeria by culture method on 596 HIV-positive Anti-Retroviral Therapy (ART) naive shows the prevalence of *Salmonella typhi* was 8.9%. Males were 242 and females were 354 (Ya'aba *et al.,* 2019).

A Hospital-based cross-sectional study conducted at Saint Paul's Hospital, Addis Ababa from December 1, 2013, to March 31, 2014, by culture method on 162 HIV seropositive Co-trimoxazole taking patients presented with diarrhea shows the prevalence of *Salmonella paratyphi* was 4.9% and *Salmonella typhi* was 3.1% (Tadesse *et al.,* 2020).

An institution-based cross-sectional study conducted at Arba Minch on 180 HIV patients from March 1 to August 31, 2019; by culture method shows that the prevalence of *Salmonella* species was 2.1%. A female was 96. A considerable proportion (38.3%) of these individuals was in the age range of 35–44 years (Ayele *et al.,* 2020).

A Hospital-based cross-sectional study conducted at Hawassa on 215 HIV patients from February to May 2016 using the culture method shows the prevalence of *Salmonella* species was 5.1%. The participants' mean age was 30 years, with ranges of 18–55 years. A female was 52% (Kebede *et al.,* 2017).

A health facility-based cross-sectional study conducted at Dessie on 112 HIV infected and 242 HIV-negative diarrheic outpatients from January 2018 to March 2018 by culture method shows *Salmonella* species prevalence among HIV-infected patients was 5.4% and non-infected was 4.5%. Their age ranged from 15 to 86 years with a mean age of 35 (Belay *et al.,* 2020).

An institution-based cross-sectional conducted at Dilla University Referral Hospital on 422 HIV participants attending the ART clinic from March to August 2022 by culture on XLD, shows prevalence of *Salmonella* species was 5.2%. The mean age of the study participants was 27.4 (±15.6 SD) years(Mitiku *et al.,* 2023).

### 2.2. Magnitude of *Shigella* species in people living with HIV

A cross-sectional study conducted in India on 45 HIV (27 with diarrhea (study group) and 18 without diarrhea (control group) included in the three-month survey of culture shows that the prevalence of *Shigella flexneri* was 2.2 %. Males were 33 and females were 12. The ages ranged from 13 - 62 years with a mean age of 34 (Shah *et al.,* 2016).

A prospective study conducted at a tertiary care Hospital in northern India between January 2014 and December 2015 on 300 HIV seropositive cases with diarrhea (study group) and 600 HIV-negative diarrhea cases (control group) shows that the prevalence of *Shigella* species was 16 (5.3%) among HIV. Out of the three hundred HIV seropositive subjects recruited, 206 (68.67%) were males and 94 (31.33%) were females. The mean age of the participants in our study was 30.64 ± 13.47 years, and the age ranged between 1 and 65 years (Goel *et al.,* 2018).

A cross-sectional study conducted at an Iraq Hospital on 70 HIV patients from 2020 to 2021 by culture method shows the prevalence of *Shigella* species was 7.14%. Age ranges between 15 to 55 years(Othman *et al.,* 2022).

A Hospital-based prospective study conducted in Nigeria from April to July 2015 by culture method on 596 HIV-positive Anti-Retroviral Therapy (ART) naive shows the prevalence of *Shigella* species was 7.9%. Males were 242 and females were 354. The mean age of the study participants was 38.0 years(Ya'aba *et al.,* 2019).

A Hospital-based cross-sectional study conducted at Saint Paul's Hospital, Addis Ababa from December 1, 2013, to March 31, 2014, on 162 HIV seropositive Co-trimoxazole taking patients

presented with diarrhea shows prevalence of *Shigella* species was 6.8%. The median age was 36 years (Tadesse *et al.*, 2020).

An institution-based cross-sectional conducted at Arba Minch on 180 HIV patients from March 1 to 31 August 2019 by culture method shows that the prevalence of *Shigella* species was 1.1%. A female was 96. The age range of 35–44 years(Ayele *et al.*, 2020).

A Hospital-based cross-sectional study conducted at Hawassa from February to May 2016 on 215 HIV-infected individuals attending the antiretroviral therapy clinic of Hawassa University Hospital shows the prevalence of *Shigella* species was 1.3%. The participants' mean age was 30.3 years with an age range of 18–55 years. The most common age group represented was those aged 45 to 54 years (34.8%). Of the study participants, 52% were females (Kebede *et al.*, 2017).

An institution-based cross-sectional conducted at Dilla University Referral Hospital among HIV Infected Patients with Diarrhea Attending the ART Clinic on 422 adult patients from March to August 2022 shows that prevalence of *Shigella* species was 7.3%. Females were 51.7%. The mean age of the study participants was 27.4 (±15.6 SD) years (Mitiku *et al.*, 2023).

### 2.3. Antimicrobial susceptibility patterns of *Salmonella* species among HIV patients

An observational study conducted in Mozambique from April 1, 2016, to February 28, 2019, by culture method on 808 HIV patients by using the Kirby Bauer diffusion method, and CLSI 2018 shows that *Salmonella* species was resistant to Penicillin (88%), Ampicillin (88%), and Amoxicillin-clavulanate (88%), and Co-trimoxazole (60%). Susceptible to Ciprofloxacin (100%), Gentamicin (100%) (Kenga *et al.,* 2021). Another cross-sectional was conducted in Mozambique on a total of 129 of which 68 were HIV and 61 were Non-HIV patients from November 2021 to May 2022 by using culture media of Salmonella-Shigella agar (Condalab, Spain), the Kirby-Bauer disk diffusion method, Mueller-Hinton agar shows *Salmonella* species were resistant to Co-trimoxazole (89.9%), Erythromycin (88.9%) and Tetracycline (76.8%). Multidrug resistance (MDR) was observed in 79.8% of *Salmonella* species (Hlashwayo *et al.,* 2023).

A cross-sectional study conducted at Cameron from September 2016 to June 2017 on 104 HIV patients using the Kirby–Bauer method (CSLI 2017) shows that Salmonella species were susceptible to Imipenem (100%), Norfloxacin (100%), and Ciprofloxacin (100%). They were also resistant to Amoxicillin-clavulanic acid (40%), Cefixime (20%), Ceftriaxone (20%), and Chloramphenicol (20%) (Ngalani *et al.*, 2019).

A cross-sectional study conducted at Nigeria Hospital on 39 HIV patients by using the culture method, (CLSI, 2018), Mueller-Hinton agar, Bauer's, and Kirby's disc diffusion method shows isolates of non-typhoid *Salmonella* were resistant to Fluoroquinolone; Nalidixic acid(100%), Augmentin(100%), Gentamicin(100%), Ofloxacin(100%), Sparfloxacin(100%), Erythromycin(100%), Chloramphenicol(100%), Ceftriaxone and Ciprofloxacin. Sensitive to Streptomycin (100%), which is a non-fluoroquinolone(Sahal *et al.*, 2020).

A Hospital-based cross-sectional study conducted at Saint Paul's Hospital, Addis Ababa from December 1, 2013, to March 31, 2014, by culture method on 162 Seropositive diarrhea patients shows *S.typhi* was resistant to Co-trimoxazole (100%), Ampicillin (100%), Nitroforantine (100 %) and Nalidixic acid (100 %), Chloramphenicol( 80%). Sensitivity of *S. para typhi* for Ceftriaxone

(62.5%). Standard Modified Kirby Bauer Disc Diffusion method and Mueller-Hinton agar medium were used(Tadesse *et al.,* 2020).

A Hospital-based cross-sectional study conducted at Hawassa on 215 HIV patients from February to May 2016 by culture method using Kirby-Bauer disc diffusion (CLSI, 2011), and Mueller-Hinton agar shows that the majority of *Salmonella* isolates were sensitive to Norfloxacin(100), Nalidixic acid, Gentamicin, Ceftriaxone, and Ciprofloxacin. Resistant to Co-trimoxazole (72%) and Chloramphenicol (72%) (Kebede *et al.,* 2017).

An institution-based cross-sectional study conducted at Arba Minch on 180 HIV patients from March 1 to 31, August 2019, by culture method using the Kirby–Bauer disc diffusion technique and (CLSI 2017 ) shows that Multidrug resistance for *Salmonella* species was 60%. Resistance of the *Salmonella* isolates to Erythromycin(80%), Ceftazidime(60%), Ampicillin(40%), Chloramphenicol (40%), Co-trimoxazole (40%), Gentamicin (40%), Tetracycline (40%), Ceftriaxone (40%), and Ciprofloxacin (20%)(Ayele *et al.,* 2020).

A cross-sectional study conducted at Dessie on 112 HIV infected from January to March 2018 by culture method using Kirby-Bauer disk diffusion, (CLSI 2017), and Muller-Hinton agar shows *Salmonella* species were resistant to Amoxicillin (100%), Ampicillin (100%), Tetracycline (47%), Co-trimoxazole (41%), Chloramphenicol (35.2%), and Nalidixic acid (17.6%). Sensitive to Ceftriaxone (100%) and Ciprofloxacin (100%)(Belay *et al.,* 2020).

An institution-based cross-sectional conducted at Dilla University Referral Hospital among HIV infected patients with diarrhea attending the ART clinic on 422 adult patients from March to August 2022 by culture method using Kirby-Bauer disk diffusion, (CLSI 2019), and Muller-Hinton agar shows *Salmonella* species were resistant to Ampicillin (80%), Co-trimoxazole (81.8%), and Ciprofloxacin (40%)(Mitiku *et al.,* 2023).

### 2.4. Antimicrobial Susceptibility Patterns of *Shigella* species among HIV patients

A cross-sectional study conducted at the Bafoussam Regional Hospital, Cameroon, from September 2016 to June 2017 on 104 HIV patients by using Kirby–Bauer method, (CSLI 2017) shows *Shigella* isolates were susceptible to Norfloxacin (100%), and Ciprofloxacin (100%). Resistant to Amoxicillin (85.71%) (Ngalani *et al.,* 2019).

A cross-sectional study was conducted in Mozambique on a total of 129 of which 68 HIV and 61 Non-HIV patients from November 2021 to May 2022 by using culture on Shigella agar (Condalab, Spain), Kirby-Bauer disk diffusion method, Mueller-Hinton II agar shows *Shigella* species resistant to Co-trimoxazole (86.6%), and Tetracycline (68.9%). Multidrug resistance was observed in 57.8% of *Shigella* species. All *Shigella* species are susceptible to Gentamicin(100%)(Hlashwayo *et al.,* 2023).

A Hospital-based cross-sectional study conducted at Saint Paul's Hospital, Addis Ababa from December 1, 2013, to March 31, 2014, on 162 HIV by culture method by using standard Modified Kirby Bauer Disc Diffusion, Mueller-Hinton agar medium shows of *Shigella* species was sensitive to Chloramphenicol(100%), Ceftriaxone(90.9%). Resistance to Co-trimoxazole (90.9%), Ampicillin (81.8%), Nitroforantione (27.3%), Nalidixic Acid (36.4%), Ceftriaxone (9.1%) Ciprofloxacin (18.%)(Tadesse *et al.,* 2020) .

An institution-based cross-sectional study conducted at Arba Minch on 180 HIV patients from March 1 to August 31, 2019 by culture method using the Kirby–Bauer disc diffusion technique and CLSI 2017 shows that Multidrug resistance was detected in *Shigella* species (100%). *Shigella* species show resistance to Ampicillin (100%), Gentamicin (100%), Erythromycin (100%), and Ceftazidime (100%). In addition, (50%) resistance to four antibiotics Tetracycline (50%), Ciprofloxacin (50%), Co-trimoxazole (50%), and Ceftriaxone (50%). Susceptible to Chloramphenicol (100%), Doxycycline (100%), Azithromycin (100%), and Meropenem (100%)(Ayele *et al.,* 2020).

A health facility-based cross-sectional study conducted at Dessie on 112 HIV-infected patients from January 2018 to March 2018 by culture method, using Standard Kirby-Bauer disk diffusion, (CLSI 2017), Muller-Hinton agar, and XLD agar shows *Shigella* species isolates were resistant to Ampicillin (100%), Co-trimoxazole (46%), and Amoxicillin (100%). Multidrug resistance of *Shigella* species (85.7%). Susceptible to Ciprofloxacin (95.6%) and Ceftriaxone (100%) (Belay *et al.*, 2020) .

An institution-based cross-sectional conducted at Dilla University Referral Hospital among HIV infected patients with diarrhea attending the ART clinic on 422 adult patients from March to August 2022 shows that *Shigella* species were sensitive to Co-trimoxazole(90.3%), Ciprofloxacin (54%), Erythromycin(96.7%), and Meropenem (96.8%). However, resistant to Ampicillin (61%)(Mitiku *et al.*, 2023).

## 2.5. Factors associated with *Salmonella* species in people living with HIV

### 2.5.1. Socio-demographic factor

#### 2.5.1.1. Age

A cross-sectional study at Desse on 112 HIV patients from January 2018 to March 2018 shows that the factor associated with non-typhoidal *Salmonella* species was the age group of 15–24 years by multivariable analysis, p-value 0.00 (Belay *et al.,* 2020). No association with the age group of 15–24 years according to a cross-sectional study, conducted at Arba Minch on 180 HIV patients from March 1 to 31 August 2019 with a p-value of 0.55 (Ayele *et al.,* 2020), and also a cross-sectional study conducted at Hawassa on 215 HIV patients from February to May 2016 shows no association with the age of 15–24 years with Crude odds ratio (at 95% CI) of 1 (Kebede *et al.,* 2017).

#### 2.5.1.2. Educational status

A cross-sectional study at Desse on 112 HIV patients from January 2018 to March 2018 shows a factor associated with non-typhoidal *Salmonella* isolates was the educational status of illiterates with a p-value of 0.04(Belay *et al.,* 2020). No association with Illiterate educational status according to an institution-based cross-sectional study conducted at Arba Minch on 180 HIV patients from March 1 to August 31, 2019, with a Crude Odd Ratio (95% CI) of 2.083 (0.7–6.21), and p-value of 0.19 (Ayele *et al.,* 2020). A cross-sectional study conducted at Hawassa on 215 HIV patients from February to May 2016 shows no association between Illiterate educational status with Crude odds ratio (95% CI) of 1 (Kebede *et al.,* 2017).

### 2.5.2. Environmental factor

#### 2.5.2.1. Source of drinking water

A health facility-based cross-sectional study conducted at Dessie on 112 HIV patients from January to March 2018 by culture method shows that a factor associated with *Salmonella* species

was drinking river or spring water sources [AOR = 8.641, 95%CI: (1.983, 37.656), P = 0.004](Belay *et al.*, 2020). Another institution-based cross-sectional study conducted at Arba Minch on 180 HIV patients from March 1 to August 31, 2019 by culture method shows that a factor associated with *Salmonella* species infection was drinking unprotected water sources (AOR 3.8, 95% CI 1.07–13.4, p 0.04)(Ayele *et al.*, 2020). No association according to a cross-sectional study conducted at Hawassa on 215 HIV patients from February to May 2016, with drinking unprotected water, with Crude odds ratio (95% CI) of 2.02 (0.69–5.96)(Kebede *et al.*, 2017).

2.5.3. Behavioral factor

2.5.3.1 Habit of hand washing after toilet

An institution-based cross-sectional study conducted at Arba Minch on 180 HIV patients from March 1 to August 31, 2019 showed that a factor associated with *Salmonella* species was lack of hand washing after toilet use (AOR 8.67, 95% CI 4.2–17.93, p 0.000). Bivariate with a P-value <0.25 and multivariable <0.05 was used (Ayele *et al.*, 2020).

2.5.3.2. Consumption of uncooked food or raw food

A Hospital-based cross-sectional study conducted at Hawassa on 215 HIV patients from February to May 2016 by culture method shows that consumption of uncooked food or raw food was significantly associated with *Salmonella* species, with a crude odds ratio of 3.41 (CI 1.13–10.3). Binary logistic regression and a 95% confidence interval were used (Kebede *et al.*, 2017). No association with the consumption of uncooked food according to an institution-based cross-sectional conducted at Arba Minch on 180 HIV patients from March 1 to 31 August 2019 with a Crude Odd Ratio of 4.15 at (95% CI) (1.13–15.24) p-value of 0.032, Adjusted Odd Ratio of 3.5 (95% CI), (0.87–14.03), p-value of 0.08 (Ayele *et al.*, 2020).

2.5.4. Clinical factor

2.5.4.1. CD4 count

A cross-sectional study conducted at Arba Minch on 180 HIV patients from March 1 to August 31 2019, shows that the factor associated with non-typhoidal *salmonella* was CD4 T-cell counts<200 cells/μL at (AOR 9.55, 95% CI 1.54–59.3, p 0.015) (Ayele *et al.,* 2020). No association according to a cross-sectional study conducted at Dessie on 112 HIV patients from January 2018 to March 2018 with non-typhoidal *Salmonella* isolates with CD4 T-cell counts<200 cells/μL with COR of 3.778 at 95% C.I(0.339–42.154), a p-value of 0.280, AOR of 1.974 at 95%CI (0.24–12.254), a p-value of 0.749. P-value < 0.05 was used (Belay *et al.,* 2020).

2.5.5. Economic factor

2.5.5.1. Absence of Toilet

A health facility-based cross-sectional study conducted at Dessie on 112 HIV patients from January to March 2018 shows a factor associated with *Salmonella* was not utilizing latrine [AOR = 6.407, 95%C.I(1.139, 36.024), P = 0.035]. P-value <0.05 was used (Belay *et al.,* 2020). No association with the use of toilets according to a cross-sectional study conducted at Arba Minch on 180 HIV patients with a Crude odds ratio of 1.49 at 95% C.I (0.52–4.32), P-value of 0.461 (Ayele *et al.,* 2020). No association with the availability of private latrines according to a cross-sectional study conducted at Hawassa on 215 HIV patients from February to May 2016 by culture method with crude odds ratio of 2.07 (0.41–10.52). Binary logistic regression and a 95% confidence interval were used (Kebede *et al.,* 2017).

2.5.5.2. Presence of domestic animal

An institution-based cross-sectional study conducted at Arba Minch on 180 HIV patients from March 1 to August 31, 2019, by culture method shows that a factor associated with *Salmonella* species was the presence of domestic animals (AOR 6.7, 95% CI 1.63–27.4, p 0.08). Bivariate with a P-value <0.25 and multivariable <0.05 was used (Ayele *et al.,* 2020).

## 2.6. Factors associated with *Shigella* species in people living with HIV

### 2.6.1. Environmental factor

#### 2.6.1.1. Source of drinking water

An institution-based cross-sectional study conducted at Arba Minch on 180 HIV patients from March 1 to August 31, 2019 shows that a factor associated with *Shigella* species was drinking unprotected water sources (AOR 3.8, 95% CI 1.07–13.4, p 0.04)(Ayele *et al.,* 2020). No association with the water source for drinking according to a cross-sectional study at Addis Ababa from December 1, 2013, to March 31, 2014, on 162 HIV patients with *Shigella* infection(Tadesse *et al.,* 2020).

### 2.6.2. Behavioral factor

#### 2.6.2.1. Habit of hand washing after toilet

An institution-based cross-sectional study conducted at Arba Minch on 180 HIV patients from March 1 to August 31, 2019 shows that a factor associated with *Shigella* species was lack of washing hands after toilet (AOR 8.67, 95% CI 4.2–17.93, p 0.000). 53.3% (n= 96) were female (Ayele *et al.,* 2020).

### 2.6.3. Clinical factor CD4 count

An institution-based cross-sectional study conducted at Arba Minch on 180 HIV patients from March 1 to August 31, 2019 shows that the factor associated with *Shigella* species was $CD_4$ less than 200 cells/μL counts( aOR 9.55, 95% CI 1.54–59.3, p 0.015)(Ayele *et al.,* 2020). No association with $CD_4$ less than 200 cells/μL counts according to a cross-sectional study conducted at Hawassa, 215 HIV patients from February to May 2016 with a COR of 1 (Kebede *et al.*, 2017). No association with $CD_4$ T-cell counts<200 cells/μL according to a cross-sectional study at Desse on 112 HIV patients with *Shigella* COR (95% C.I) of 3.778 (0.339–42.154), a p-value of 0.280, AOR (95%CI) of 1.974 (0.24–12.254), a p-value of 0.749(Belay *et al.*, 2020).

## 2.7. Conceptual framework

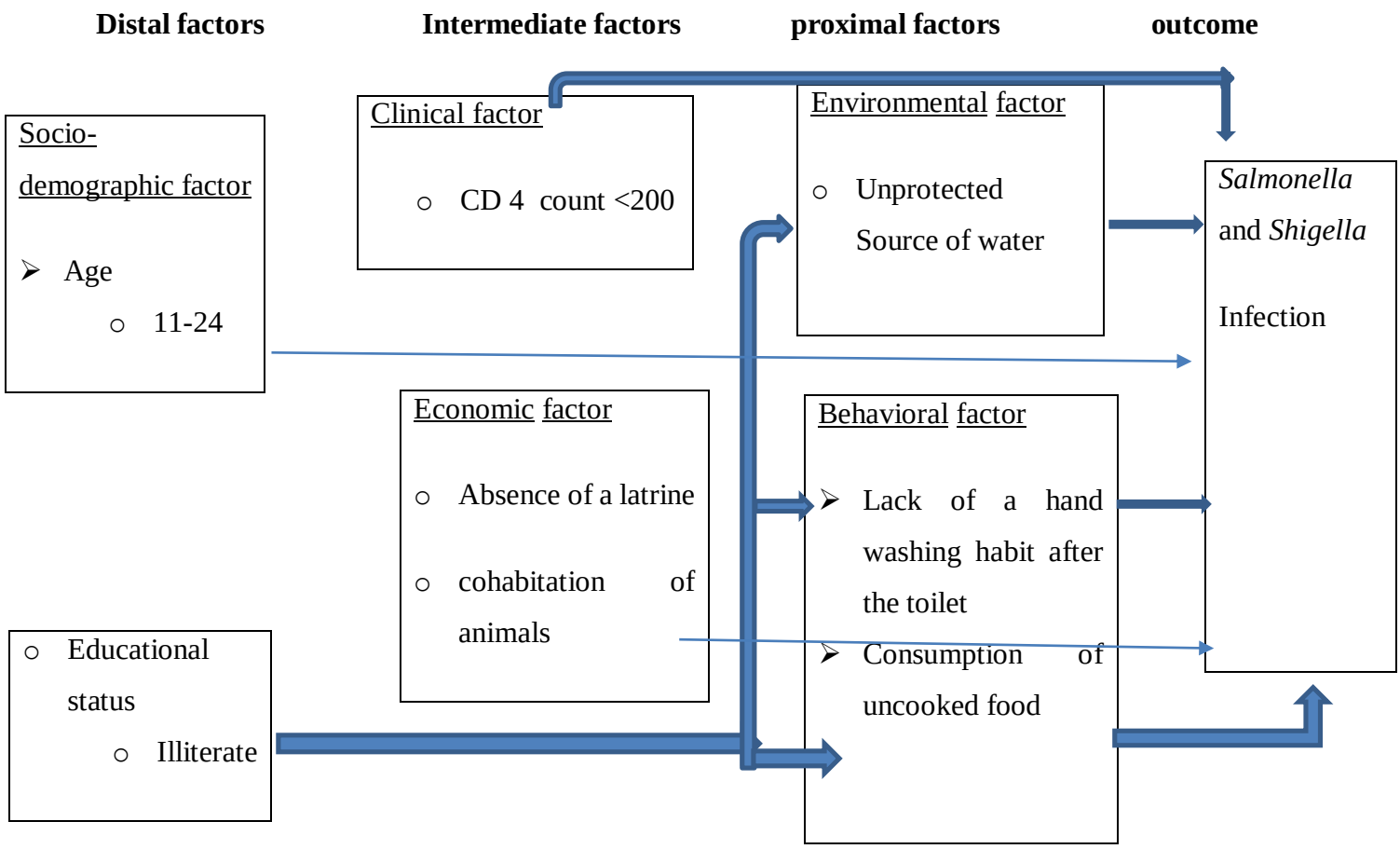


**Figure 1.** Conceptual framework on factors associated with *Salmonella* and *Shigella species* among HIV patients. (Source: constructed by the investigator from reading different literature)

# 3. MATERIALS AND METHODS

## 3.1. Study area and period

This study was conducted among HIV patients attending ART treatment at Hiwot Fana Comprehensive Specialized University Hospital, Harar, Ethiopia from February 01 to April 30, 2024. Harar is the capital city of the Harari regional state, located 525 km east of Addis Ababa, the capital city of Ethiopia. The population in Harar for 2022 is 151,977 (Projection of 2022 Based on the 2007 Census conducted by the Central Statistical Agency of Ethiopia). Harar is one of 40 cities in Ethiopia and ranks 11th in the Ethiopian population, has 9 Districts (6 urban and 3 rural), and 36 kebeles (Statistics of Population of Harar in 2023).

There are seven Hospitals in the Harari Region of which one is owned by the Harari Regional Health Bureau while the rest are either other governmental or other private Hospitals. Among these, the 2 Hospitals are governmental public health facilities. There are also 8 public health centers, 32 health posts, 10 not-for-profit private clinics, and 15 private clinics for profit in the Harari Region((Shama *et al.*, 2021);*(*Harari Regional Health Bureau report)).

Hiwot Fana Compressive Specialized University Hospital provides healthcare services to more than 5 million people around Harar and neighboring regions like Eastern Oromia, Dire Dawa City Administration, and Somali Regional State. They are delivering different health services to the community like surgery, dental care, ophthalmologist, internal medicine, gynecology and obstetrics, pediatrics, Maternal and Child Health (MCH), TB and HIV (TB/HIV), intensive medical care, mental health care, dermatology, and venereal disease services, pharmacy, oncologist, and various laboratory services and so on. The Hospital has 1121 workers; from these 512 are health workers, 71 General practitioners (GP), 100 Specialists, and 438 Administration workers(Source: Hiwot Fana Human Resource Management 2023 Records). Currently, the facility is treating more than 1920, of this 1302 was females and male was 618. This Hospital was selected because it gives service to most HIV patients in the region.

## 3.2. Study design

An institutional-based cross-sectional study was conducted among HIV patients**.**

## 3.3. Populations

### 3.3.1. Source population

All HIV-positive patients who were on ART drugs and naïve HIV patients with diarrhea attended treatment at Hiwot Fana Comprehensive Specialized University Hospital, Harar, Ethiopia.

### 3.3.2. Study population

HIV-positive patients who were on ART and naïve HIV patients with diarrhea attended treatment during the study period at Hiwot Fana Comprehensive Specialized University Hospital, Harar, Ethiopia.

## 3.4. Inclusion and exclusion criteria

### 3.4.1. Inclusion criteria

HIV-infected individuals with diarrhea were included during the study period at Hiwot Fana Comprehensive Specialized University Hospital, Harar, Ethiopia.

### 3.4.2. Exclusion criteria

HIV Patients who took antibiotics in the last 2 weeks, who were critically ill and unable to conduct an interviewed, or unable to provide stool were excluded from the study.

### 3.5. Sample size determination

**The sample size determination for objective 1**: A single population formula was used considering the magnitude of *Salmonella* species at Dessie 5.4% (Belay *et al.,* 2020), the margin of error of (0.05) or 5%, d=0.05, and Z=1.96. By $n=Z^2P(1-P)/d^2 = (1.96)^2 0.054(1-0.054)/0.0025$ = 1.96*1.96*0.054*0.946*/0.0025 = 0.1962/0.0025 =78 by considering 10% non-response our sample size was 86.

**The sample size determination for objective 2:** A single population formula was used considering the magnitude of *Shigella* species at Addis Ababa 6.8%(Tadesse *et al.,* 2020), margin of error of (0.05) or 5% d=0.05, P=prevalence(6.8%), and Z=1.96. By $n= (1.96)^2 0.068(10.068)/0.0025$= 1.96*1.96*0.068*0.932/0.0025 =97 HIV patient. By considering 10% non-response, our sample size was 107.

**The sample size determination for objective 3**: Sample size for factors associated with *Salmonella* species among HIV patients by using a double population proportion formula. The sample size was calculated for the associated factors obtained from different literature by using statics of Epi Info statistical software version 7.2 with an assumption confidence level of 95%, a power =80%, and a ratio of unexposed to exposed almost equivalent to 1.

**Table 1:** Sample size determination for a factor associated with *Salmonella* species among HIV patients.

| Factors | The outcome in exposed (%) | The outcome in unexposed (%) | Sample size with 10% non-response | Reference |
|---|---|---|---|---|
| Drinking river or spring water | 25 | 2.4 | 96 | (Belay *et al.,* 2020) |
| Consumption of raw food | 65.6 | 34.4 | 52 | (Kebede *et al.,* 2017) |

**The sample size determination for objective 4**: Sample size for a factor associated with *Shigella* species among HIV patients by using the double population proportion formula. The sample size was calculated for the associated factors obtained from different literature by using statics of Epi Info statistical software version 7.2 with the assumption of a confidence level=95%, power =80%, and the ratio of unexposed to exposed almost equivalent to 1.

**Table 2:** Sample size determination for a factor associated with *Shigella* species among HIV patients.

| Factor for *Shigella* | The outcome in exposed (%) | The outcome in unexposed (%) | Sample size with 10% non-response | Reference |
|---|---|---|---|---|
| unprotected sources of drinking water | 40 | 60 | 235 | (Ayele *et al.*, 2020) |
| Consumption of raw food | 65.6 | 34.4 | 52 | (Kebede *et al.*, 2017) |

Sample size was sample size for the magnitude of *Salmonella* species 86 (Belay *et al.*, 2020), the sample size for the magnitude of *Shigella* 107(Tadesse *et al.*, 2020), the sample size for the associated factor of *Salmonella* 96(Belay *et al.*, 2020), the sample size for the associated factor of *Shigella* species 235(Ayele *et al.*, 2020).

The **final sample size** for the study was by taking the maximum sample size, which is **235 HIV patients** attending ART treatment at Hiwot Fana Compressive Specialized Hospital (Ayele *et al.*, 2020).

### 3.6. Sampling techniques

Convenient sampling techniques were used to include 235 HIV patients with complaint of diarrhea who are attending treatment at Hiwot Fana Comprehensive Specialized University Hospital.

### 3.7. Method of Data Collection

#### 3.7.1. Data collector

Three data collectors (nurses) collected the socio-demographic data using questionnaires and clinical data (Level of CD4 count, WHO clinical stage, adherence to HIV drugs) from the medical record.

#### 3.7.2. Data Collection Instrument

Data was collected by using a pretested structured questionnaire. A questionnaire was first prepared in the English language and then translated into the local language (Amharic and Afaan Oromoo) and back to the English language. Data was collected using face-to-face interviews with structured questionnaires by the data collector( three nurses) about socio-demographic variables (age, sex, educational status, marital status, occupational status, residence, family size), environmental factor (source of drinking water), behavioral factors (hand washing habit after toilet, consumption of raw food or uncooked food, habit of eating raw meat, habit drinking raw milk, hand washing before meal, habit of raw vegetable and fruit), and economic factors (availability of toilets or latrine, presence domestic of animals). Clinical factors (Level of $CD_4$ count, WHO clinical stage, adherence to HIV drugs) from medical records.

#### 3.7.3. Data Collection Procedure

Data was collected through a face-to-face interview with their local language to collect the socio-demographic data. The data collector also collected clinical data from medical records.

#### 3.7.4. Stool sample collection, handling, and Transport

Instruction was given to study participants on how to collect the sample by principal investigator at microbiology laboratory. Two grams of semi-formed stool, mucoid and bloody stool or 2 ml watery stool sample was collected from the study participants using a coded disposable plastic cup and processed on the same day.

### 3.7.5. Culture and Bacterial Identification

Stool samples were inoculated on selenite F broth (Oxoid, Hampshire, UK) at 35°C–37°C for 18–24 hours (Cheesbrough, 2005), Then sub cultured on MacConkey agar (Liofilchem Italy), and Xylose Lysine Deoxycholate (XLD; Oxoid) agar incubated at 35°C–37°C for 18–24 hours. After 24 hours of incubation, nonlactose fermenter on MacConkey and the colony on XLD media was evaluated for the presence of bacterial growth (Cheesbrough, 2005). The culture plates were examined for the presence of *Salmonella* species (Pink–red with black center colonies or red colony) and *Shigella* species (Pink-red colonies). All suspected colonies were inoculated onto appropriate biochemical test including Indole test(MIO, HIMEDIA, India), Citrate test(Liofilchem, Italy), Triple sugar iron test(Himedia, India), Lysine decarboxylase test(LIA, Liofilchem, Italy), and Urease test Christensen( Liofilchem, Italy). The result of each biochemical was read after incubation for 24–48 hours at 37 °C(Cheesbrough, 2005). Colonies producing an alkaline slant with acid butt, and hydrogen sulfide production on Triple Sugar Iron Agar, positive for Lysine decarboxylase, negative for urea hydrolysis, negative for indole test, positive for citrate utilization and motility test were considered as *Salmonella* species. Whereas, urease negative, indole positive/negative, pink-red slope and yellow production in butt with no blackening in Triple Sugar Iron agar, Lysine decarboxylase negative, non-motile and citrate negative was considered as *Shigella* species(Cheesbrough, 2005).

### 3.7.6. Antimicrobial susceptibility testing

The antimicrobial susceptibility test was done using the modified Kirby-Bauer disk diffusion techniques on Mueller–Hinton agar (Biomark, India) by the Clinical and Laboratory Standards Institute guideline (CLSI 2020). About 3–5 colonies of the same type were mixed with 5 ml of sterile normal saline (0.85% NaCl). The density of the suspension was determined by visual comparison with 0.5 McFarland standard. A sterile cotton swab was used to distribute the bacterial suspension evenly over the entire surface of the Mueller–Hinton agar plates. The inoculated plates were left at room temperature for 3-5 minutes to dry. Antimicrobial disks were applied to the surface of the inoculated plates using sterile forceps. Antimicrobial disks including Ampicillin (A-

10 µg), Tetracycline (TE-30 mg), Chloramphenicol (C-30 µg), Gentamicin (GM-10 µg, Ciprofloxacin (CIP-5 µg), Trimethoprim-sulfamethoxazole (1.25/23.75 µg), Nalidixic acid (NA-30 µg), Amoxicillin-clavulanate (20/10 µg), Ceftriaxone (CRO-30 µg) and Azithromycin (ATH-15 µg) was applied on the culture plates (CLSI 2020). Selection based on CLSI 2020, an operational definition of XDR *Shigella* and *Salmonella*, guidelines of the Hospital treatment and different literature reviews. The zone of inhibition was read after 24 hours of incubation at 37 °C by ruler. The results were recorded as sensitive (S), resistant (R), or intermediate (I) based on (CLSI 2020). Isolated bacteria were preserved in Tryptic soy Broth at -20 °C (Liofilchem, Italy).

### 3.8. Study variable

#### 3.8.1. Dependent variable

*Salmonella* and *Shigella* isolates

#### 3.8.2. Independent variables

Socio-demographic variables (age, sex, educational status, marital status, occupational status, residence, family size)

Environmental factor (source of drinking water)

Behavioral factors (hand washing habit after toilet, consumption of raw food or uncooked food, habit of eating raw meat, habit of drinking raw milk, hand washing before meal, habit of eating raw vegetables and fruit)

Clinical factors (Level of $CD_4$ count, WHO clinical stage, adherence to HIV drug treatment)

Economic factors (Availability of toilets, cohabitation of animals)

### 3.9. Operational Definitions

**Diarrhea** is the condition of having loose, watery stools occurring three or more times a day. It is often accompanied by urgency, abdominal cramps, and dehydration in severe cases(Levine *et al*., 2017).

**Extreme Drug Resistance (XDR) typhoid** is resistant to at least five antibiotic classes recommended to treat typhoid fever, including Ampicillin, Ceftriaxone, Chloramphenicol, Ciprofloxacin, and Co-trimoxazole (Trimethoprim-Sulfamethoxazole) CDC (Centers for Disease Control and Prevention)(Medalla *et al.,* 2021).

**Extreme Drug Resistance (XDR) *Shigella* bacteria: -** CDC defines XDR *Shigella* bacteria as strains that are resistant to all commonly recommended empiric and alternative antibiotics such as Azithromycin, Ciprofloxacin, Ceftriaxone, Co-trimoxazole(Trimethoprime-Sulphamethoxazole), and Ampicillin (Centers for Disease Control and Prevention) (Waller)2023).

**Intermediates (I):-** a category defined by zone diameters within the intermediate range or for which the response rate may be lower than for susceptible isolates(CLSI, 2020).

**Multi-drug-resistant:** Resistance to at least 1 antibiotic from at least 3 separate antibiotic classes*(*Rafailidis and Kofteridis, 2022).

**Naïve HIV patients: -** HIV-positive patients who has never taken antiretroviral drugs (Dean, 2021).

**Resistant(R):-** a category defined by zone diameter at or below the resistant break point that is not inhibited by the usually achievable concentrations of the agent with normal dosage schedules(CLSI, 2020).

**Sensitive(S)** is defined by zone diameter at or above the susceptible breakpoint that is inhibited by the usually achievable concentrations of an antimicrobial agent when the dosage recommended to treat the site of infection is used resulting in likely clinical efficacy (CLSI, 2020).

### 3.10. Data quality control

After preparing the questionnaire, a pretest was done on 10% of the study sample size of HIV patients at Jugol General Hospital before the actual data collection for the validity and reliability of the questionnaire. During pretesting, additional information was gathered, and unclear terms, phrases, and questions were identified and given modifications. The training was provided for the data collectors (three nurses) on the data collection tools and sample collection procedure before the actual sample collection started. The completeness of the questionnaire was checked on daily bases whether necessary information was fulfilled properly. During the processing of each sample standard operational procedure (SOPs) was followed. To check the sterility of the culture media, 5% of the media was incubated without inoculation. The sterility of the material was checked by indicator paper for culture media sterility. The instrument and reagent were checked for reliability and reproducibility of the test before starting to examine by using known positive and negative samples. Culture media quality was checked, and sterilized, and the Anti-microbial sensitivity test expiration date was checked. Quality control for culture media, biochemical test, and the Anti-microbial sensitivity test was checked by positive control and negative control. The quality of each new batch of culture medium and antimicrobial disks was checked by testing *E. coli* (ATCC 25922) and *Salmonella typhimurium* (ATCC 14028) which was obtained from HFCSUH. Day to Day-to-day quality was checked for the presence of growth or changes in culture media growth.

### 3.11. Method of Data Processing and Analysis

The collected data was checked for its completeness, coded, and entered into Epi-Data version 3.1, and it was analyzed by using the Statistical Package for Social Science (SPSS) version 26. Descriptive statistics (mean, percentages, and standard deviation) were calculated to summarize results and present the results in words, charts, graphs, and tables. Logistic regression (bivariate and multivariable) was performed to check the presence of an association between the dependent variable with the independent variables and associated factors were analyzed by SPSS statistics. In bivariate analysis, variables with a P-value of less than 0.25 were considered a candidate for

multivariable analysis. In multivariable analysis variables with P-values less than 0.05 at a 95% confidence interval were considered statistically significant.

### 3.12. Ethical consideration

Ethical clearance was obtained from the Institutional Health Research Ethics Review Committee (IHRERC) of the College of Health and Medical Sciences, Haramaya University (Ref.no.IHRERC/155/2023. Official letters of support were written to Hiwot Fana Comprehensive Specialized University Hospital (HFCSUH) (ሀ/ለ/ሳ/100/1486). The study was explained to the study participants and the head of the Hospital before data collection. Voluntary, informed, written, and signed consent was obtained from each selected study participant and head Hospital. For age 15-18 assent was obtained from parents. The study participants were informed of their right to refuse or withdraw from the interview at any time. Any information and findings obtained during the study were kept confidential by excluding names and identifiers in the questionnaire. Individuals infected with *Salmonella* or *Shigella* were treated according to antibiotic susceptibility.

### 3.13. Information Dissemination

The first copy of this study was submitted to Haramaya University, College of Health and Medical Sciences, Hiwot Fana Comprehensive Specialized Hospital, Regional Health Bureau, and other interested groups. In addition, the finding of the research was presented in a seminar, and publication was attempted in scientific journals as well.

# 4. RESULTS

## 4.1. Sociodemographic characteristics

This study was conducted on 235 HIV positive individuals on ART who completed interview and stool examination, showing a response rate of 91.5 %. The mean age of the study participants was 42.5 (SD ± 11.7 years), median age of 44.0, and a range of 15 to 78 years. The majority of the study participants were in the age group 41- 45 years (21.7%), female (75.7%), lived in urban (84.7%), and married (46.8%). The higher number of study participants had primary school (1–8) by educational status (45.1%); and housewife (27.2%) by occupational status. The average family size of the study participants was 3.30 with +/- 1.76 standard deviation. Most study participants had 1-3 (57.9%) family size (Table 3).

**Table 3:** Socio-demographic profile of HIV Patients with complaints of diarrhea at HFCSUH, Harar Ethiopia, 2024 (N = 235)

| Variables | Category | Frequency | % |
|---|---|---|---|
| Age | 15-20 | 16 | 6.8 |
| | 21 -25 | 10 | 4.3 |
| | 26-30 | 14 | 6.0 |
| | 31-35 | 14 | 6.0 |
| | 36-40 | 40 | 17.0 |
| | 41-45 | 51 | 21.7 |
| | 46-50 | 44 | 18.7 |
| | 51-55 | 20 | 8.5 |
| | 56-60 | 17 | 7.2 |
| | 61-65 | 5 | 2.1 |
| | 66-70 | 1 | 0.4 |
| | 71-75 | 1 | 0.4 |
| | 76-78 | 2 | 0.9 |

| | | | |
|---|---|---|---|
| Sex | Male | 58 | 24.7 |
| | Female | 177 | 75.7 |
| Residence | Urban | 199 | 84.7 |
| | Rural | 36 | 15.3 |
| Education level | No formal education | 48 | 20.4 |
| | Primary school (1–8) | 106 | 45.1 |
| | Secondary school (9–12) | 55 | 23.4 |
| | College/University | 26 | 11.1 |
| Occupation | Gov't employee | 51 | 21.7 |
| | Housewife | 64 | 27.2 |
| | Merchant | 40 | 17.0 |
| | Student | 22 | 9.4 |
| | Farmer | 8 | 3.4 |
| | Other(daily labor) | 50 | 21.3 |
| Marital status | Single | 37 | 15.7 |
| | Married | 110 | 46.8 |
| | Divorced | 44 | 18.7 |
| | Widowed | 44 | 18.7 |
| Family size | 1-3 | 136 | 57.9 |
| | 4-6 | 89 | 37.9 |
| | >6 | 10 | 4.3 |

### 4.2. Magnitude of *Salmonella* and *Shigella* species

The overall magnitude of *Salmonella* and *Shigella* species among HIV-infected individuals was observed to be 7.7 %( 18/235) (95% CI: 4.0-11.0). *Salmonella* species 4.7% (11/235) (95% CI: 2.0-7.0) and *Shigella* 3% (7/235) (95% CI: 1.0-5.0).

### 4.3. Factors associated with the *Salmonella* and *Shigella* species among HIV Patients with complaints of diarrhea

In the bivariate analysis, place of residence, educational status, family size, un protected source of water, hand wash after toilet use, hand wash before meal, drinking raw milk, eating raw meat, raw vegetable, presence of domestic animals in house, adherence to drug, WHO clinical stage, and $CD_4$<200 were the candidate variables for multivariable analysis.

In multivariable analysis, unprotected sources of water, and clinical stage3 and 4 were statistically significant with p values ⩽0.05). HIV patients drinking from Unprotected water sources were 8.10 times more likely to be infected with *Salmonella* and *Shigella* infection than those using protected water sources (AOR=8.1; 95% CI: 1.54-42.61) (Table 4)

**Table 4:**Factors associated with the prevalence of *Salmonella* and *Shigella species* among HIV patients attending ART treatment at Hiwot Fana Comprehensive Specialized University Hospital, Harar, Ethiopia, from February 01 to April 30, 2024 (N = 235).

| Variables | Category | Proportion of *Salmonella* and *Shigella* species | | COR (95% CI) | p-value | AOR (95% CI) | p-value |
|---|---|---|---|---|---|---|---|
| | | Positive (%) | Negative (%) | | | | |
| Age(In year) | <35 | 6(10.9) | 49(89.1) | 1 | | | |
| | ≥35 | 12(6.7) | 168(93.3) | 0.58 (0 .21-1.64) | 0.305 | | |

| | | | | | | | |
|---|---|---|---|---|---|---|---|
| Sex | Male | 6 (10.3) | 52(89.7) | 1 | | | |
| | Female | 12(6.8) | 165(93.2) | 1.59(0.57 - 4.44) | 0.379 | | |
| Residence | Urban | 11 (5.5) | 188(94.5) | 1 | | 1 | |
| | Rural | 7(19.4) | 29(80.6 ) | 4.13 (1.48 - 11.49) | 0.007 | 1.93(0.21-17.69) | 0.559 |
| Educational status | Not read and write | 6(12.2) | 43(87.8) | 2.02 (0.72 - 5.68) | 0.182 | 0.22(0.03-1.93) | 0.171 |
| | Literate | 12(6.5) | 174(93.5) | 1 | | 1 | |
| Occupation | Employed | 6(11.3) | 47(88.7) | 1 | | | |
| | Non employed | 12(6.6) | 170(93.4) | 0.55 (0.19 - 1.55 ) | 0.260 | | . |
| Marital status | Single, widowed, Divorced | 8(6.8) | 117(93.2 ) | 1 | | | |
| | Married | 10(9.1) | 100(90.9) | 1.46 (0.56 -3.85) | 0.441 | | |
| Family size | <4 | 8 (5.8) | 129(94.2) | 1 | | 1 | |
| | ≥4 | 10(10.2 ) | 88(89.8 ) | 1.83 (0.69 - 4.83 ) | 0.220 | 1.81(0.31-10.59) | 0.510 |
| Source of drinking water | Protected (tap water only) | 6(3.2) | 184(96.2) | 1 | | 1 | |
| | Unprotected | 12(26.7) | 33(73.3 ) | 11.15 ( 3.91 - 31.79) | 0.000 | 8.10(1.54-42.61) | 0.013 * |
| Latrine accessibility | Yes | 7(6.7) | 97(93.3) | 1 | | | |
| | No | 11(8.4 ) | 120(91.6 ) | 1.27 (0.48-3.40) | 0.634 | | |
| Hand washing after the use of the toilet | Yes | 10(4.7 ) | 205(95.3) | 1 | | 1 | |
| | No | 8(40) | 12(60) | 13.67(4.56-40.93) | 0.000 | 6.18(0.71-54.13) | 0.100 |

| | | | | | | | |
|---|---|---|---|---|---|---|---|
| Hand wash before meals | Yes | 12(5.5) | 207(94.5) | 1 | | 1 | |
| | No | 6(37.5) | 10(62.5) | 10.35(3.22-33.27) | 0.000 | 5.88(0.81-42.51) | 0.079 |
| Eat uncooked food | Yes | 9 (7.3) | 115(92.7 ) | 0.89 (0.34-2.32) | 0.810 | | |
| | No | 9(8.1) | 102(91.9 ) | 1 | | 1 | |
| Consumption of raw milk | Yes | 9(17.6) | 42(82.4 ) | 4.17 (1.56 - 11.14) | 0.004 | 2.06(0.36-11.85) | 0.420 |
| | No | 9(4.9 ) | 175(95.1) | 1 | | 1 | |
| Consumption of raw meat | Yes | 6(6.1) | 87(93.9) | 2.99 (1.08-8.262) | 0.035 | 3.96(0.66-23.844) | 0.132 |
| | No | 12(4.4) | 130(95.6 ) | 1 | | 1 | |
| Consumption of raw vegetables | Yes | 10(5.2) | 183(94.8 ) | 0.23 (0 .09 - 0.63) | 0.004 | 0.31(0.05-2.59) | 0.305 |
| | No | 8(19.1) | 34(80.9) | 1 | | 1 | |
| Domestic animals in the house | Yes | 11(17.2) | 53(82.8) | 4.86 (1.79 - 13.18 ) | 0.002 | 1.19(0.19-7.67) | 0.853 |
| | No | 7(4.1) | 164(95.9 ) | 1 | | 1 | |
| Adherence to drug | Good | 12 (5.5) | 207(94.5) | 1 | | 1 | |
| | Poor | 6(37.5) | 10(62.5 ) | 10.35 (3.22-33.27 ) | 0.000 | 4.52(0.54-38.00) | 0.165 |
| WHO clinical stage | Stage1 and 2 | 9(4.3) | 200(95.7) | 1 | | 1 | |
| | Stage3 and 4 | 9(34.6) | 17(65.4) | 11.77 (4.12-33.56 ) | 0.000 | 27.27(14.13-40.41) | 0.001 * |
| Current CD4+ (cells/µl) | <200 | 6(26.1) | 17(73.9) | 5.88(1.96-17.64) | 0.002 | 0.13(0.01-1.20) | 0.072 |
| | ≥200 | 12(5.7) | 200(94.3) | 1 | | 1 | |

*p<0.05

### 4.5. Antibiotic susceptibility pattern of HIV Patients with a complaint of diarrhea at HFCSUH, Harar Ethiopia, 2024 (N = 18).

All *Salmonella* isolates (n=11) and *Shigella* isolates (n=7) were 100% susceptible to Ciprofloxacin. About 85.7 % and 90.1% *Shigella* and *Salmonella* isolates were susceptible to Ceftriaxone, respectively. The highest resistance was shown by isolates of *Salmonella* species against Ampicillin 11(100%), Nalidixic acid 6(54.5%), Cotrimoxazole5 (45.5%) and Chloramphenicol 4(45.5%) while low resistance was observed against Ceftriaxone 1(9.9%), to Gentamycin 2 (18.2%), and Amoxicillin-Claviculamic AMC 2(18.2%). *Salmonell*a isolates showed 1(9.9%) intermediate susceptibility against Amoxicillin-Claviculamic (AMC). *Shigella* species shows intermediate susceptibility to Chloramphenicol 1(14.3%). The highest resistance was shown by isolates of *Shigella* species against three antibiotics tested Ampicillin 7 (100%) Tetracycline6 (85.7%), and Nalidixic acid 5(71.4). and Co-trimoxazole 4(57.1%), lower resistance to Gentamicin 2(28.6%), Amoxicillin-claviculamic acid 1(14.3%), Ceftriaxone 1(14.8%), and Ciprofloxin 0 percentage) (Table 5).

**Table 5:** Antimicrobial susceptibility pattern of *Salmonella* and *Shigella* species isolated from HIV patients with diarrhea at HFCSUH, Harar, Ethiopia, 2024 (N = 18).

| Bacteria | | Antibiotics n (%) | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|
| *Shigella* species | | CIP | CRO | AMP | SXT | AMC | CAF | AZTH | GEN | TTC | NAL |
| | S | 7(100%) | 6(85.7%) | - | 3(42.9%) | 6(85.7%) | 3(42.3% | 5(71.4%) | 5(71.4%) | 1(14.3%) | 2(28.6%) |
| | I | - | - | - | - | - | 1(14.3%) | - | - | - | - |
| | R | - | 1(14.8%) | 7(100%) | 4(57.1%) | 1(14.3%) | 3(42.3%) | 2(28.6) | 2(28.6%) | 6(85.7%) | 5(71.4) |
| *Salmonella* species | S | 11(100%) | 10(90.1%) | - | 6(54.5%) | 8(72.7%) | 6(54.5%) | 7(63.6) | 9(81.8%) | 7(63.6%) | 5(45.5%) |
| | I | - | - | - | - | 1(9.9%) | - | - | - | - | - |
| | R | - | 1(9.9%) | 11(100%) | 5(45.5%) | 2(18.2%) | 5(45.5%) | 4(36.4%) | 2(18.2%) | 4(36.4%) | 6(54.5%) |
| Total (n = 18) | S | 18(100%) | 16(88.8%) | - | 9(50%) | 14(77.7) | 9(50%) | 12(66.7%) | 14(77.8) | 8(44.4%) | 7(38.9%) |
| | I | - | - | - | - | 1(5.6%) | 1(5.6%) | | - | - | - |
| | R | - | 2(11.1) | 18(100%) | 9(50%) | 3(16.6%) | 8(72.7%) | 6(33.33%) | 4(22.2%) | 10(55.6%) | 11(61.1%) |

Abbreviations: R Resistant, I Intermediate, S Sensitive, Key: AMP Ampicillin, TTC Tetracycline, GEN Gentamicin, NAL Nalidixic acid, CRO ceftriaxone, SXT Trimethoprime-Sulphamethoxazole (co-trimoxazole), CAF Chloramphenicol, CIP Ciprofloxacin, AMC amoxicillin-clavulanic AZTH Azithromycin

### 4. 6. Multidrug resistance pattern of HIV Patients with a complaint of diarrhea at HFCSUH, Harar Ethiopia, 2024 (N = 18).

The overall multidrug resistance of *Salmonella* and *Shigella* species was 50 %( 9/18). Of these, 54.5 %( 6/11) were *Salmonella* species and *Shigella* species 42.8 %( 3/7). Resistance to two antibiotics resistance *Salmonella* and *Shigella* species was 27.7 %( 5/18). Of this, 27.3 %( 3/11) was *Salmonella* species and 28.6 %( 2/7) *Shigella* species. Resistance to 1 antibiotic for *Salmonella* and *Shigella* species was 22.2 %( 4/18). Of this, 27.3 % (3/11) was *Salmonella* species, and *Shigella* species 14.3 %( 1/7). The most frequent MDR was observed in *Salmonella* isolates 54.5 %( 6/11) (Table 6).

**Table 6:** Antibiogram of *Shigella* and *Salmonella* isolated among HIV patients attending ART treatment at Hiwot Fana Comprehensive Specialized University Hospital, Harar, Ethiopia, from February 01 to April 30, 2024

| **Antimicrobial combination** | *Salmonella* No (%) N=11 | *Shigella* No (%) N=7 | Total No % N=18 |
|---|---|---|---|
| Ampicillin | 3(27.3%) | 1(14.3%) | 4(22.2%) |
| Ampicillin and Nalidic acid | 1(9.1%) | | 1(5.6%) |
| Ampicillin and Tetracycline | 1(9.1%) | 2(28.6%) | 3(37.5%) |
| Ampicillin and Co-trimoxazole | 1(9.1%) | | 1(5.6%) |
| AMP,SXT,AZTH | 1(9.1%) | | 1(5.6%) |
| AMP,TETRA,NAL | | 1(9.1%) | 1(5.6%) |
| AMP,NAL,GEN,CAF, | 1(9.1%) | | 1(5.6%) |
| AMP,SXT,CAF,AZTH | 1(9.1%) | | 1(5.6%) |
| AMP,NAL,TTC,CAF,SXT | | 1(14.3%) | 1(5.6%) |
| AMP,SXT,CAF,TTC,NAL | | 1(14.3%) | 1(5.6%) |
| AMP,SXT,CAF,AZTH,TTC,NAL | 1(9.1%) | | 1(5.6%) |
| AMP,AMC,CAF,AZTH,GEN,NAL | | 1(14.3%) | 1(5.6%) |
| AMP,SXT,CAF,AZTH,GEN,TTC,NAL | 1(9.1%) | | 1(5.6%) |

Abbreviations Key: AMP Ampicillin, TTC Tetracycline, GEN Gentamicin, NAL Nalidixic acid, CRO ceftriaxone, SXT Trimethoprime-Sulphamethoxazole (co-trimoxazole), CAF Chloramphenicol, CIP Ciprofloxacin, AMC amoxicillin-clavulanic AZTH Azithromycin

# 5. DISCUSSION

In this study, the overall prevalence of *Salmonella* and *Shigella* species was 7.7 % (95% CI: 4.0-11.0). This was comparable with studies conducted, in Ethiopia at Hawassa 6.4% (Kebede *et al.,* 2017), at Dilla 10.5%(Mitiku *et al.,* 2023) and in India (8.3 %) (Goel *et al.,* 2018). However, it was higher than the study conducted at Arba Minch 3.2 %( Ayele *et al.,* 2020) but Lower than study conducted at Addis Ababa (11.7%) (Tadesse *et al.,* 2020) and Iraq(19.9%)(Othman *et al.,* 2022).

In this study, the magnitude *Shigella* species was 3 %( 95% CI: 1.0-5.0). This is similar to study conducted at Arba Minch (1.1%) (Ayele *et al.,* 2020), and at Hawassa (1.3%) (Kebede *et al.,* 2017). The current finding is lower than studies conducted in Ethiopia at Dilla (7.3%) (Mitiku *et al.,* 2023), Addis Ababa (6.8%) (Tadesse *et al.,* 2020), Nigeria (7.9%) (Ya'aba *et al.,* 2019) and Iraq (7.14%) (Othman *et al.,* 2022).

The current study detected *Salmonella* species in 4.7 % (95% CI: 2.0-7.0) HIV-infected diarrhea patients. This finding was in line with study in Ethiopia at Dessie 5.4% (Belay *et al.,* 2020), at Hawassa 5.1% (Kebede *et al.,* 2017) , at Arba Minch 2.1%(Ayele *et al.,* 2020), Cameron (2.9% )(Mabeku *et al.,* 2020), at India ( 5.3%) (Goel *et al.,* 2018), and However; our finding was lower than studies at Iraq (12.8%) (Othman *et al.,* 2022) and at Mozambique (11%)(Kenga *et al.,* 2021).

The difference in the distribution of *Salmonella* and *Shigella* might be due to geographical variation and socioeconomic conditions, awareness about of health promotion, and methodological differences (sample size, study design, study period, and diagnostic techniques, poor personal hygiene, and environmental sanitation(Senthilkumar *et al.,* 2014) and local meteorology (Dallal *et al.,* 2020) and seasonal (Lee *et al.,* 2017) (Marriott *et al.,* 2018). For instance, a study conducted in Iraq (12.8%) found that geographical variation is responsible for the high magnitude of *Salmonella* infection(Othman *et al.,* 2022). Another study conducted in Nigeria (7.9%) socioeconomic elements factors responsible for high magnitude of *Shigella* infection (Ya'aba *et al.,* 2019).

The study's data implicate impact for HIV patients, *Salmonella* and *Shigella* infections can be significantly more severe, leading to more prolonged and severe symptoms like diarrhea, fever, and abdominal pain compared to individuals with healthy immune systems, due to the compromised immune function caused by HIV, which increases the risk of complications like bacteremia (bacteria entering the bloodstream) and potentially life-threatening infections; essentially, even a typical case of *Salmonella* or *Shigella* can become a serious medical concern for someone with HIV(Dikman *et al.,* 2015) .

Our study shows among empirical treatment used to treat *Salmonella* species resistance to Trimethoprime-Sulphamethoxazole (Co-trimoxazole) (45.5%). This is consistent with reports from Arba Minch (40%) (Ayele *et al.,* 2020), and at Dessie (41%)(Belay *et al.,* 2020). Higher reports from Mozambique (60%)(Kenga *et al.,* 2021), another in Mozambique(89.9%)(Hlashwayo *et al.,* 2023), and at Hawassa (72%)(Kebede *et al.,* 2017). *Shigella* species resistant to Co-trimoxazole for this study was (57.1%). This is consistent with Arba Minch (50%)(Ayele *et al.,* 2020). Lower at Dessie (46%), (Belay *et al.,* 2020) . Higher reports from Addis Ababa (90.9%) (Tadesse *et al.,* 2020) ,and at Mozambique (86.6%)(Hlashwayo *et al.,* 2023).

In the current study, all *Shigella* species were resistant to Ampicillin (100%). This finding was comparable with the study conducted in Ethiopia ; at Arba Minch (100%)(Ayele *et al.,* 2020), and at Dessie (100%) (Belay *et al.,* 2020). Lower resistance was reported study conducted in Ethiopia :Addis Ababa (81.8%)(Tadesse *et al.,* 2020), and Dilla (61%)(Mitiku *et al.,* 2023). The resistance rate of *Salmonella* species was 100% to Ampicillin. This finding was comparable with studies conducted at Mozambique (88%) (Kenga *et al.,* 2021) in different parts of Ethiopia, at Dessie(100%) (Belay *et al.,* 2020), lower at Arba Minch(40%)(Ayele *et al.,* 2020).

This study shows *Salmonella* species resistance to Tetracycline was 36.4%. This is consistent with reports from Ethiopia : at Arba Minch (40%)(Ayele *et al.,* 2020), and at Dessie (47%)(Belay *et al.,* 2020). Higher reports from Mozambique (76.8%)(Hlashwayo *et al.,* 2023). The resistance of *shigella* to Tetracycline (85.7%). This result was higher than the studies reported from Mozambique (68.9%)(Hlashwayo *et al.,* 2023), and at Arba Minch (50%)(Ayele *et al.,* 2020).

This study shows *Salmonella* species resistance to Chloramphenicol (45.5%). This is consistent with reports at Dessie (35.2%) (Belay *et al.,* 2020), and Arba Minch (40%) (Ayele *et al.,* 2020). Lower reports from Cameron (20%) (Ngalani *et al.,* 2019). However, resistance of *Shigella* species to Chloramphenicol was (42.3%). This is consistent with reports at higher report from lower resistance or no resistance at Addis Ababa (Tadesse *et al.,* 2020), and Arba Minch (Ayele *et al.,* 2020).

The reason for the variation of Antimicrobial resistance for above mentioned antimicrobial resistance is may be due to the overuse and misuse of antibiotics in the humans (Morehead and Scarbrough, 2018), the nature of the susceptibility of strains the immune status of the study participants, geographic region, syndromic diagnosis and diagnostic imprecision, poor infection control in health care settings, poor hygiene and sanitation, absence of new antibiotics being discovered((Tang *et al.,* 2023); (Morehead and Scarbrough, *2018*); (Erku *et al.,* 2017); (Castro-Sánchez *et al.,* 2016*)*).

This study implicates antimicrobial resistance patterns stresses the importance of surveillance programs to monitor antimicrobial trends. This is critical for updating clinical guidelines and managing emerging threats in resource-limited settings

In this study, Multi-Drug Resistance (MDR) resistance of *Salmonella* species was 54.5%. This is comparable to the study conducted at Arba Minch ( 60%) (Ayele *et al.,* 2020) and , lower than reports from Mozambique (79.8%) (Hlashwayo *et al.,* 2023). While the current study reported MDR of *Shigella* species was 42.8%. This findings is lower than studies conducted in other parts of Ethiopia such as Arba Minch (100%)(Ayele *et al.,* 2020), Dessie ( 85.7%) and at Mozambique ( 57.8%)(Hlashwayo *et al.,* 2023).

The cause of variations in the prevalence of MDR is unknown but might be due to inappropriate empirical antimicrobial treatment, easy availability, and indiscriminate use of common antimicrobials(Tang *et al.,* 2023) . The current Ethiopian standard guide treats HIV patient's initial Ciprofloxacin for both *Salmonella* and *Shigella*, Alternatives Cotrimoxazole, Ceftriaxone, Cefotaxime ((of the Infectious *et al.,* 2022*)*; (AIDSinfo, 2019)). The use Cotrimoxazole should be

according to antimicrobial susceptibility test because resistance high at this study area. This study implies for clinical management and treatment the urgent need for tailored treatment strategies based on antimicrobial susceptibility testing rather than empirical treatments.

*Salmonella* and *Shigella* was known transmitted by draining of feacally contaminated water(Liu *et al.,* 2018) . In this study, *Salmonella* and *Shigella* species infections associated with unprotected source of water. This is similar to studies conducted in Ethiopia : at Dessie (Belay *et al.,* 2020), and Arba Minch(Ayele *et al.,* 2020). However , at Addis Ababa, Ethiopia study reported ,there is no association between the type of water source for drinking with *Salmonella* and *Shigella infection* (Tadesse *et al.,* 2020). The variation is may be due to difference in protection of source of drinking water. For instance in study conducted at Addis Ababa majority of the study participants drinks protected water (Tadesse *et al.,* 2020).

The second factors identified in the current study was clinical stages 3 and 4. Even though clinical stage of HIV patients were not studied by previous literature. The clinical stage of HIV infection is related to the risk of *Salmonella* and *Shigella* infection because HIV weakens the immune system, making the body more susceptible to infection*(*Rameshkumar and Arunagirinathan, 2018*)*.

### 5.1. Strength and Limitation of study

*Salmonella* and *Shigella* was identified to genus level using culture and biochemical test. The methods used antimicrobial susceptibility pattern of *Salmonella* and *Shigella* species are also comprehensive. This study possibly applied to study population in the study area. However, Characterization of the isolates to species level was not done due to a lack of serological tests. As the study was cross-sectional, it does not establish a temporal relationship between cause and effect. This study was conducted only on patients presented with diarrhea cases and not include asymptomatic /sub clinical carrier which can under estimate the magnitude report

# 6. CONCLUSIONS AND RECOMMENDATIONS

## 6.1. Conclusions

This study revealed a 7.7% prevalence of *Salmonella* and *Shigella* species among HIV patients in the study area, which is relatively high. Both species are developing resistance to the commonly prescribed antibiotic like Ampicillin. *Salmonella* and *Shigella* isolates were sensitive to Ciprofloxacin. There were also higher drug resistance and multidrug-resistant patterns. *Salmonella* and *Shigella* infections were significantly associated with unprotected water and clinical factors.

## 6.2. Recommendation

**To health professionals and Hospital:**

- Clinician should have to consider culture-based bacterial species identification and antimicrobial susceptibility testing services for treatment *Salmonella* and *Shigella* infections. Thus Hospitals should work to establishment of culture and AST facilities.
- Ciprofloxacin can be used as the empiric first-line antibiotics in any suspected case of *Salmonella* and *Shigella* species among HIV patients. Ampicillin as an empiric first-line antibiotic in the management of suspected should be discouraged
- Health information dissemination to HIV patients about transmission by focusing unsafe water and stage 3 and 4 clinical stage patients.
- Health information decimation to raise awareness HIV patients of antimicrobial resistance and appropriate used antimicrobials

**Harari regional health bureau:**

- Further studies on status of water supply and provision of safe water supply.
- Establishment of culture-based bacterial species identification and antimicrobial susceptibility testing services at health facility setting and also enhanced surveillance to evaluate these trends.

**To researcher:**

- Continuous surveillance of the prevalence and antibiotic resistance pattern of *Salmonella* and *Shigella* species among HIV patients in different setting by including large sample size, different factors and intestinal parasites.
- However, the study verified that further studies should be conducted on patients attending health centers and private clinics. The economic status of study participants was not studied for this study.

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# 8. ANNEXES

## 8.1. Information Sheet and Informed Voluntary Consent Form for HFCSUH

**1. Introduction:** My name is Sisay Geremew Gurmu. I am the principal Investigator of a study to be conducted in your Hospital for my Master's degree at Haramaya University, College of Health and Medical Sciences. I kindly request you to lend me your attention to explain to you about the study and your institution being selected as the study setting.

**2. The study title:** To determine the magnitude, associated factors, and antimicrobial susceptibility patterns of *Salmonella* and *Shigella* species among Human Immunodeficiency Virus patients attending treatment at Hiwot Fana Comprehensive Specialized University Hospital, Harar, Ethiopia.

**3. Purpose of the study:** The finding of this study can be of paramount importance for Hiwot Fana Comprehensive Specialized University Hospital to treat HIV patients' according to drug susceptibility patterns when they are infected by *Salmonella* and *Shigella* infection. Moreover, this study aims to write a thesis as a partial requirement for the fulfillment of a Master's Program in microbiology for the principal investigator.

**4. Procedure and duration:** I was interviewing patients using a questionnaire to provide me with pertinent data that is helpful for the study. There are 18 questions to answer and I will fill the questionnaire by interviewing HIV patients. The interview with each HIV patient will take about 18 minutes. A stool sample is needed at the end of the questionnaire.

**5. Risks and benefits**: The risk of participating in this study is very minimal, but only takes a few minutes from patients' time. There would not be any direct payment for participating in this study. However, the finding from this research will help Hiwot Fana Comprehensive Specialized University Hospital to treat HIV patients according to their drug susceptibility patterns.

**6. Confidentiality**: The information that we was provided was kept confidential. There was no information that will identify the participants in particular. The findings of the study was general

for the study participants and will not reflect anything particular about individual persons. The questionnaire will be coded to exclude showing names. No reference will be made in oral or written reports that could link participants to the research.

**7. Rights**: Participation in this study is fully voluntary. The participants have the right to declare whether to participate or not in this study. If they decide to participate, they have the right to withdraw from the study at any time and this will not label them for any loss of benefits, to which are entitled. They have the right not to answer any question that they do not want to answer. The institution has the right to know about the study and for a logical reason the right not to accept this consent form.

**8. Contact address**: If there are any questions or inquiries at any time about the study or the procedures, please contact: Address of principal investigator: Name: Sisay Geremew Gurmu, Mobile number 0940770023/0921316710, E-mail address: Sisaygaramu83@gmail.com. Address of Institutional Health Research Ethics Review Committee (IHRERC): Office phone: 0254662011 P.O.BOX: 235, Harar, Ethiopia.

**9. Declaration of informed voluntary consent**: I have read the participant information sheet. I have clearly understood the purpose of the research, the procedures, the risks and benefits, issues of confidentiality, the rights of participating, and the contact address for any queries. I have been allowed to ask questions about things that may have been unclear. I understand that participant has the right to withdraw from the study at any time or not to answer any question that they do not want. I am also informed that the Hospital has the right to stop this study from being conducted if any misdeeds and unethical procedures are observed during the data collection process on the Hospital's premises. Therefore, I declare my volunteer on behalf of the Hiwot Fana Comprehensive Specialized University Hospital Management to allow this study to be conducted in our Hospital with my signature as indicated below.

Name and Signature of Head of the Hospital: _____ _____ Date _____

Name and Signature of the Principal Investigator: _____ _____ Date _____

**Thank you for your cooperation**

**8.2. Participant Information Sheet and Informed Voluntary Consent Form (for competent adults: ages ≥18 years)**

**1. Introduction:** My name is----------.I am working as a data collector for the study being conducted in Hiwot Fana Comprehensive Specialized University Hospital by Sisay Geremew Gurmu who is studying for his master's degree at Haramaya University, College of Health and Medical Sciences. I kindly request you to lend your attention to explain to you about the study and being selected as the study participant.

**2. The study title:** To determine the magnitude, associated factors, and antimicrobial susceptibility patterns of *Salmonella* and *Shigella* species among Human Immunodeficiency Virus patients attending treatment at Hiwot Fana Comprehensive Specialized University Hospital, Harar, Ethiopia.

**3. Purpose of the study:** The finding of this study can be of paramount importance for Hiwot Fana Comprehensive Specialized University Hospital to treat HIV patients' according to drug susceptibility patterns when they are infected by *Salmonella* and *Shigella* infection. Moreover, this study aims to write a thesis as a partial requirement for the fulfillment of a Master's Program in microbiology for the principal investigator.

**4. Procedure and duration:** I will be interviewing you using a questionnaire to provide pertinent data that is helpful for the study. There are 18 questions to answer and I will fill the questionnaire by interviewing you. The interview will take about 18 minutes, so I kindly request you spare me this time for the interview. You will give a stool sample at the end of the questionnaire.

**5. Risks and benefits:** The risk of participating in this study is very minimal, but only taking a few minutes of your time. There would not be any direct payment for participating in this study. But the findings from this research may help Hiwot Fana Comprehensive Specialized University Hospital to treat HIV patients with appropriate drug treatment when they are infected with *Salmonella* or *Shigella* species.

**6. Confidentiality**: The information you will provide us will be confidential. There was no information that will identify you in particular. The findings of the study was general for the study participants and will not reflect anything particular about individual persons or housing. The questionnaire will be coded to exclude showing names. No reference will be made in oral or written reports that could link participants to the research.

**7. Rights:** Participation in this study is fully voluntary. You have the right to declare whether to participate or not in this study. If you decide to participate, you have the right to withdraw from the study at any time and this will not label you for any loss of benefits to which you otherwise are entitled. You do not have to answer any question that you do not want to answer.

**8. Contact address:** If there are any questions or enquires any time about the study or the procedures, please contact the Address of the principal investigator: Name: Sisay Geremew Gurmu, Mobile number 0940770023/0921316710, E-mail address: Sisaygaramu83@gmail.com. Address of Institutional Health Research Ethics Review Committee (IHRERC): Office phone 0254662011 P.O.BOX: 235, Harar, Ethiopia.

**9. Declaration of informed voluntary consent:** I have read/ was read to me the participant information sheet. I have clearly understood the purpose of the research, the procedures, the risks and benefits, issues of confidentiality, the rights of participating, and the contact address for any queries. I have been given the opportunity to ask questions for things that may have been unclear. I was informed that I have the right to withdraw from the study at any time or not to answer any question that I do not want. Therefore, I declare my voluntary consent to participate in this study with my initials (signature).

Name and signature of participant: _____ _____ Date _____

Name and signature of Data Collector: _____ _____ Date _____

**Thank you for your cooperation**

**8.3. Participant information sheet and informed voluntary assent obtained from parents form for a minor (age < 18 years)/vulnerable individual to be signed by his/her legal competent representative (e.g.: -parent/guardian)**

**1. Introduction:** My name is----------. I am working as a data collector for the study being conducted in Hiwot Fana Comprehensive Specialized University Hospital by Sisay Geremew Gurmu who is studying for his master's degree at Haramaya University, College of Health and Medical Sciences. Your child is randomly selected to be a participant in this study. I kindly request you to lend me your attention to explain to you about the study and the child's participation.

**2. The study title:** To determine magnitude, associated factors, and antimicrobial susceptibility patterns of *Salmonella* and *Shigella* species among Human Immunodeficiency Virus patients attending treatment at Hiwot Fana Comprehensive Specialized University Hospital, Harar, Ethiopia.

**3. Purpose of the study:** The finding of this study can be of paramount importance for Hiwot Fana Comprehensive Specialized University Hospital to treat HIV patients' according to drug susceptibility patterns when they are infected by *Salmonella* and *Shigella* infection. Moreover, this study aims to write a thesis as a partial requirement for the fulfillment of a Master's Program in microbiology for the principal investigator.

**4. Procedure and duration:** I will interview your child using a questionnaire to provide me with pertinent data that is helpful for the study. I will ask 18 questions to answer and I will fill the questionnaire by interviewing. The interview will take about 18 minutes. A stool sample is needed at the end of the questionnaire. Therefore, I kindly request you spare me this time and allow me to perform this procedure on your child.

**5. Risks and benefits:** The risk of participating for your child in this study is very minimal, but only taking a few minutes of your time. There would not be any direct payment for participating in this study. But the findings from this research may help Hiwot Fana Comprehensive Specialized

University Hospital to treat HIV patients with appropriate drug treatment when they are infected with *Salmonella* or *Shigella* species.

**6. Confidentiality:** The information that we will collect from this study will be confidential. There will be no information that will identify your child or yourself in particular. The findings of the study will be general for the study community and will not reflect anything particular about individual persons or housing. The data that we gather from the questionnaire will exclude showing names. No reference will be made in oral or written reports that could link participants to the research.

**7. Rights:** Participation in this study is fully voluntary. You have the right to declare whether to allow your child to be involved in this study or not. If you would allow your child for this study, you have the right to withdraw him/her from the study at any time and this will not label you/your child for any loss of benefits to which you/your child otherwise are entitled. You do not have to answer any question that you do not as well.

**8. Contact address:** If there are any questions or enquires any time about the study or the procedures, please contact: Address of principal investigator: Name: Sisay Geremew Gurmu, Mobile number 0940770023/0921316710, E-mail address: Sisaygaramu83@gmail.com. Address of Institutional Health Research Ethics Review Committee (IHRERC): Office phone: 0254662011 P.O.BOX: 235, Harar, Ethiopia.

**9. Declaration of informed voluntary consent:** I have read/ was read to me/ the participant information sheet. I have clearly understood the purpose of the research, the procedures, the risks and benefits, issues of confidentiality, the rights of participating, and the contact address for any queries. I have been given the opportunity to ask questions for things that may have been unclear. I was informed that I have the right to withdraw my child from the study at any time or not to answer any question that I do not want. Therefore, I declare my voluntary consent to allow my child to participate (be involved) in this study with my initials (signature) Name and signature of Parent/Guardian: _____ _____ Date: _____ Name and signature of Data Collector: _____ _____ Date: _____**Thank you for your cooperation**

**8.4. Amharic Version Information Sheet and Informed Voluntary Consent Form (for competent adults: ages ≥18 years)**

**1. መግቢያ፡** ስሜ፡----------በህይወት ፋና ኮምፕሬሄንሲቭ ስፔሻላይዝድ ዩኒቨርስቲ ሆስፒታል በሀሮማያ ዩኒቨርሲቲ የጤናና ህክምና ሳይንስ ኮሌጅ ሁለተኛ ዲግሪያቸውን በመማር ላይ ባለው በሲሳይ ገረመው ጉርሙ ለሚካሄደው ጥናት መረጃ ሰብሳቢ ሆኜ እየሰራሁ ነው። ስለ ጥናቱ እና የጥናቱ ተሳታፊ ሆነው መመረጣቸውን ለእርስዎ ለማስረዳት ትኩረትዎን እንዲሰጡ በአክብሮት እጠይቃለሁ።

**2. የጥናቱ ርዕስ፡** በሂወት ፋና ኮምፕረሄንሲቭ ስፔሻላይዝድ ዩኒቨርስቲ ሆስፒታል፣ ሃረር፣ ኢትዮጵያ ህክምና በመከታተል ላይ የሚገኙትን የኤችአይቪ ታካሚን የሳልሞኔላ (ታይፎይድ) እና የሺግላ መጠን፣ ተያያዥነት ያለው ምክንያት እና ፀረ-ተህዋሲያን ሁኔታ ለማወቅ።

**3. የጥናቱ ዓላማ፡** የሂወት ፋና ኮምፕረሄንሲቭ ስፔሻላይዝድ ዩኒቨርሲቲ ሆስፒታል የኤችአይቪ ታማሚዎችን በሳልሞኔላ እና በሺግላ በሽታ ሲያዙ እንደ መድሀኒት ተጋላጭነት ሁኔታ ለማከም የዚህ ጥናት ግኝት ከፍተኛ ጠቀሜታ ይኖረዋል። ከዚህም በላይ የዚህ ጥናት ዓላማ ለማስተርስ መርሃ ግብር በማይክሮባዮሎጂ ለዋናው መርማሪ ከፊል መስፈርት ሆኖ ጥናታዊ ፅሁፍ መጻፍ ነው።

**4. የአሰራር ሂደቱ እና የቆይታ ጊዜ፡** ለጥናቱ የሚረዳ ጠቃሚ መረጃ ለማቅረብ መጠይቁን ተጠቅሜ ቃለ መጠይቅ አደርግልዎታለሁ። እርስዎን በመጠየቅ መጠይቁን የምሞላባቸው 18 ጥያቄዎች መልስ ያገኛሉ። ቃለ-መጠይቁ 18 ደቂቃ ያህል ይወስዳል፣ስለዚህ ለቃለ መጠይቁ በዚህ ጊዜ እንድትቆጥቡልኝ በአክብሮት እጠይቃለሁ።

**5. ስጋቶች እና ጥቅማ ጥቅሞች፡** በዚህ ጥናት ጉዳት በጣም አናሳ ነው፣ ነገር ግን ከእርስዎ ጊዜ ጥቂት ደቂቃዎችን ብቻ ነው የሚወስደው። በዚህ ጥናት ውስጥ ለመሳተፍ ምንም አይነት ቀጥተኛ ክፍያ አይኖርም። ነገር ግን የዚህ ጥናት ግኝቶች ህይወት ፋና ኮምፕረሄንሲቭ ስፔሻላይዝድ ዩኒቨርስቲ ሆስፒታል የኤችአይቪ ታማሚ በሳልሞኔላ (ታይፎይድ) ወይም በሺጋላ ባክቴሪያ ሲጠቃ ተገቢውን የመድሃኒት ህክምና እንዲያገኝ ሊረዳው ይችላል።

**6. ምስጢራዊነት፡** እርስዎ የሚያቀርቡልን መረጃ ሚስጥራዊ ይሆናል። በተለይ እርስዎን የሚለይ መረጃ አይኖርም። የጥናቱ ግኝቶች ለጥናት ማህበረሰብ አጠቃላይ እና የግለሰብን ወይም የመኖሪያ ቤትን ምንም የሚያንፀባርቅ አይሆንም። መጠይቂያው ስሞችን ከማሳየት እንዲገለሉ ኮድ ይደረጋል። ተሳታፊዎችን ከጥናቱ ጋር ሊያገናኙ የሚችሉ የቃል ወይም የጽሁፍ ዘገባዎች ማጣቀሻ አይደረግም።

**7. መብቶች:** የዚህ ጥናት ተሳትፎ ሙሉ በሙሉ በፈቃደኝነት ነው. በዚህ ጥናት ለመሳተፍም ሆነ ላለመሳተፍ የማወጅ መብት አልዎት። ለመሳተፍ ከወሰኑ, በማንኛውም ጊዜ ከጥናቱ የመውጣት መብት አለዎት እና ይህ እርስዎ ያለዎትን ጥቅማጥቅሞች ማጣት ላይ ምልክት አያደርግልዎትም. መመለስ የማትፈልገውን ማንኛውንም ጥያቄ መመለስ የለብህም።

**8. የእውቂያ አድራሻ፡** ስለ ጥናቱ ወይም ሂደቶቹ ምንም አይነት ጥያቄዎች ካሉ ወይም በማንኛውም ጊዜ የሚጠይቅ ከሆነ እባክዎን ያነጋግሩኝ፡- ኢ-ሜል፡ (E-mail): sisaygaramu83@gmail.com, የሞባይል ቁጥር 0940770023/0921316710, የተቋማዊ ጤና ጥናትና ምርምር ስነምግባር ግምገማ ኮሚቴ አድራሻ: የቢሮ ስልክ፡ 0254662011 P.O.BOX: 235, Harar, Ethiopia.

**9. በመረጃ ላይ የተመሰረተ የፈቃደኝነት ስምምነት መግለጫ፡**-የተሳታፊውን የመረጃ ወረቀት አንብቤአለሁ/ ተነብቦልኛል። የጥናቱ ዓላማ፣ አካሄዶች፣ ስጋቶች እና ጥቅሞች፣ ሚስጥራዊ ጉዳዮች፣ የመሳተፍ መብቶች እና ለማንኛውም መጠይቆች የእውቂያ አድራሻውን በግልፅ ተረድቻለሁ። ግልጽ ባልሆኑ ጉዳዮች ላይ ጥያቄዎችን እንድጠይቅ እድል ተሰጥቶኛል። በማንኛውም ጊዜ ከጥናቱ የመውጣት ወይም የማልፈልገውን ማንኛውንም ጥያቄ ላለመመለስ መብት እንዳለኝ ተነገረኝ። ስለዚህ፣ በዚህ ጥናት የመጀመሪያ ሆኜ (ፊርማ) ላይ ለመሳተፍ የፈቃዴ ፈቃዴን አውጃለሁ።

የተሳታፊው ስም እና ፊርማ፡-_____ _____ ቀን _____

የ መረጃ  ሰብሳቢው ስም እና ፊርማ፡_____ _____ ቀን _____

**ለትብብርዎ እናመሰግናለን**

**8.5. Amharic version participant information sheet and informed voluntary assent obtained from parents form for a minor (age < 18 years)/vulnerable individual to be signed by his/her legal competent representative (e.g.: -parent/guardian**

1.***መግቢያ:*** ስሜ:----------በህይወት ፋና ኮምፕረሄንሲቭ ስፔሻላይዝድ ዩኒቨርሲቲ ሆስፒታል በሀሮማያ ዩኒቨርሲቲ የጤናና ህክምና ሳይንስ ኮሌጅ ሁለተኛ ዲግሪያቸውን በመማር ላይ ባለው በሲሳይ ገረመው ጉርሙ ለሚካሄደው ጥናት መረጃ ሰብሳቢ ሆኜ እየሰራሁ ነው። ልጅዎ በዚህ ጥናት ውስጥ እንዲሳተፍ በዘፈቀደ ተመርጧል። ስለ ጥናቱ እና የልጁ ተሳትፎ ለእርስዎ ለማስረዳት ትኩረትዎን እንዲሰጡኝ በአክብሮት እጠይቃለሁ።

**2. የጥናቱ ርዕስ፡** በሂወት ፋና ኮምፕረሄንሲቭ ስፔሻላይዝድ ዩኒቨርስቲ ሆስፒታል፣ ሃረር፣ ኢትዮጵያ ህክምና በመከታተል ላይ የሚገኙትን የኤችአይቪ ታካሚን የሳልሞኔላ እና የሺግላ መጠን፣ ተያያዥነት ያለው ምክንያት እና ፀረ-ተህዋሲያን ሁኔታ ለማወቅ።

**3.የጥናቱ ዓላማ፡** የሂወት ፋና ኮምፕረሄንሲቭ ስፔሻላይዝድ ዩኒቨርሲቲ ሆስፒታል የኤችአይቪ ታማሚዎችን በሳልሞኔላ እና በሺግላ በሽታ ሲያዙ እንደ መድሀኒት ተጋላጭነት ሁኔታ ለማከም የዚህ ጥናት ግኝት ከፍተኛ ጠቀሜታ ይኖረዋል። ከዚህም በላይ የዚህ ጥናት ዓላማ ለማስተርስ መርሃ ግብር በማይክሮባዮሎጂ ለዋናው መርማሪ ከፊል መስፈርት ሆኖ ጥናታዊ ፅሁፍ መጻፍ ነው።

**4. የአሰራር ሂደቱ እና የቆይታ ጊዜ፡-** ለጥናቱ ጠቃሚ የሆነ ጠቃሚ መረጃ እንዲሰጠኝ መጠይቁን ተጠቅሜ ከልጅዎ ጋር ቃለ መጠይቅ አደርጋለሁ። በቃለ መጠይቅ መጠይቁን የት እንደምሞላ ለመመለስ 18 ጥያቄዎችን እጠይቃለሁ። ቃለ መጠይቁ 18 ደቂቃ ያህል ይወስዳል። በመጠይቁ መጨረሻ ላይ የሰገራ ናሙና ያስፈልጋል። ስለዚህ, በዚህ ጊዜ እንድትቆጠቡኝ እና ይህን አሰራር በልጅዎ ላይ እንድፈጽም በትህትና እጠይቃለሁ።

**5. ስጋቶች እና ጥቅማ ጥቅሞች፡** በዚህ ጥናት ውስጥ ልጅዎ በመሳተፍ የሚደርስበት አደጋ በጣም አነስተኛ ነው; ግን ከእርስዎ ጊዜ ጥቂት ደቂቃዎችን ብቻ ይወስዳል። በዚህ ጥናት ውስጥ ለመሳተፍ ምንም አይነት ቀጥተኛ ክፍያ አይኖርም። ነገር ግን የዚህ ጥናት ግኝቶች ህይወት ፋና ኮምፕረሄንሲቭ ስፔሻላይዝድ ዩኒቨርስቲ ሆስፒታል የኤችአይቪ ታማሚ በሳልሞኔላ ወይም በሺጋላ ባክቴሪያዎች ሲጠቃ ተገቢውን የመድሃኒት ህክምና እንዲያገኝ ሊረዳው ይችላል።

**6. ሚስጥራዊነት፡** ከዚህ ጥናት የምንሰበስበው መረጃ ሚስጥራዊ ይሆናል። በተለይ ልጅዎን ወይም እራስዎን የሚለይ መረጃ አይኖርም። የጥናቱ ግኝቶች ለጥናት ማህበረሰብ አጠቃላይ እና የግለሰብን ወይም የመኖሪያ ቤትን ምንም የሚያንፀባርቅ አይሆንም። ከመለኪያዎቹ የምንሰበስበው መረጃ ስሞችን ማሳየትን ያስወግዳል። ተሳታፊዎችን ከጥናቱ ጋር ሊያገናኙ የሚችሉ የቃል ወይም የጽሁፍ ዘገባዎች ማጣቀሻ አይደረግም።

**7. መብቶች፡** የዚህ ጥናት ተሳትፎ ሙሉ በሙሉ በፈቃደኝነት ነው. ልጅዎ በዚህ ጥናት ውስጥ እንዲሳተፍ ወይም እንዳይሳተፍ ለመፍቀድ የመግለጽ መብት አለዎት። ልጅዎን ለዚህ ጥናት ከፈቀዱ፣ በማንኛውም ጊዜ ከጥናቱ የማውጣት መብት አልዎት እና ይህ እርስዎ/ልጃችሁ ያለዎት ጥቅማጥቅም ማጣት እርስዎ/ልጃችሁ ላይ ምልክት አይደረግም። እርስዎም ለማይፈልጉት ማንኛውንም ጥያቄ መመለስ የለብዎትም።

**8. የእውቂያ አድራሻ፡** ስለ ጥናቱ ወይም አሠራሩ ማንኛውም አይነት ጥያቄ ወይም ጊዜ የሚጠይቅ ከሆነ፡ የዋና መርማሪ አድራሻ፡ ሲሳይ ገረመው ጉርሙ ሞባይል ቁጥር 0940770023/0921316710 በኢሜል አድራሻ፡ E-mail Sisaygaramu83@gmail.com ያግኙ። የተቋማዊ ጤና ጥናትና ምርምር ሥነምግባር ገምጋሚ ኮሚቴ (IHRERC) አድራሻ፡ የቢሮ ስልክ፡ 0254662011 P.O.BOX: 235, Harar, Ethiopia.

**9. በመረጃ ላይ የተመሰረተ የፈቃደኝነት ስምምነት መግለጫ፡-**

የተሳታፊውን የመረጃ ወረቀት አንብቤአለሁ/ተነብቦልኛል/። የጥናቱ ዓላማ፣ አካሄዶች፣ ስጋቶች እና ጥቅሞች፣ ሚስጥራዊ ጉዳዮች፣ የመሳተፍ መብቶች እና ለማንኛውም መጠይቆች የእውቂያ አድራሻውን በግልፅ ተረድቻለሁ። ግልጽ ባልሆኑ ጉዳዮች ላይ ጥያቄዎችን እንድጠይቅ እድል ተሰጥቶኛል። ልጄን በማንኛውም ጊዜ ከጥናቱ የማስወጣት ወይም የማልፈልገውን ማንኛውንም ጥያቄ ላለመመለስ መብት እንዳለኝ ተነገረኝ። ስለዚህ፣ ልጄ በዚህ ጥናት የመጀመሪያ ፊደሌ (ፊርማ) እንዲሳተፍ (እንዲሳተፍ) ለመፍቀድ በፈቃደኝነት መስማማቴን አውጃለሁ።

የወላጅ/አሳዳጊ ስም እና ፊርማ፡- _____ _____ ቀን፡ _____

የመረጃ ሰብሳቢው ስም እና ፊርማ፡- _____ _____ ቀን፡_____

**ለትብብርዎ እናመሰግናለን**

**8.6. Afaan Oromo Version Information Sheet and Informed Voluntary Consent Form (for competent adults: age's ≥ 18 years)**

**1. Seensa:** Maqaan Koo----------dha. Ani qorannoo Hospitaala Ispeeshaalistii Waliigalaa Yuunivarsiitii Hiwot Faanaa keessatti Sisay Geremew Gurmu Yunivarsiitii Haramayaa, kolleejjii fayyaa fi saayinsii fayyaa keessatti digrii lammaffaa isaaf barachaa jiruuf ragaa walitti qabaa ta'uun hojjechaa jira. Waa'ee qorannichaa fi hirmaataa qorannichaa ta'ee filatamuu akkan isiniif ibsuuf xiyyeeffannoo keessan akka naaf keenitaan kabajaan isin gaafadha.

**2. Mata duree qorannichaa:** Tatamsa'ina Saalmooneelaa fi Shigeelaa, sababoota walqabataniifi haala farra maaykiroobiyaanii dhukkubsattoota HIV Hospitaala Yuunivarsiitii Ispeeshaalaayizd waliigalaa Hiwot Fana, Harar, Ethiopia keessatti yaalamaa jiran adda baasuuf.

**3. Kaayyoo qorannichaa:** Argannoon qorannoo kanaa dhukkubsattoota HIV dhukkuba Salmonella fi Shigella qaban Hospitaala Ispeeshaalaayizeeshinii Yuunivarsiitii Waliigalaa Hiwot Fana keessatti wal'aanuuf barbaachisummaa guddaa qabaata. Kana malees, kaayyoon qorannoo kanaa qorataa ijoo sagantaa maaykiroobaayoloojiitiif akka barbaachisummaa gartokkeetti barruu qorannoo barreessuu dha.

**4. Adeemsaa fi yeroo:** Odeeffannoo barbaachisaa ta'ee fi qorannichaaf gargaaru kennuu dhaaf gaaffilee fayyadamee isin gaafadha. Gaaffilee 18 deebisuuf jiran keessatti gaaffii fi deebii isiniin godheen guuta. Gaaffii fi deebii gara daqiiqaa 18 waan fudhatuuf yeroo kana gaaffii fi deebii kanaaf akka na qusattan kabajaan isin gaafadha.

**5. Balaa fi faayidaa**: Miidhaan qorannoo kana irratti hirmaachun dhufu xiqqaa dha, garuu yeroo kee keessaa daqiiqaa muraasa qofa fudhata... Qorannoon Kun hirmaachuuf kaffaltiin kallattiin hin jiru. Haa ta'u malee, argannoon qorannoo kanaa Hospitaalii Ispeeshaalistii Yuunivarsiitii Waliigalaa Hiwot Faanaa yeroo dhukkubsataan HIV baakteeriyaa Salmonellan ykn Shigellan qabamu yaala qoricha sirrii ta'e akka argatu gargaaruu danda'a.

**6. Iccitii:** Odeeffannoon isin nuuf kennitan iccitii ta'a. Odeeffannoon addatti si adda baasu hin jiraatu. Argannoon qorannichaa hawaasa qorannichaaf waliigalaa kan ta'u yoo ta'u, namoota dhuunfaa ykn mana jireenyaa adda ta'e kan hin calaqqisiifne ta'a. Gaaffiin maqaa agarsiisu akka hin dabalamneef koodii ni kennama. Gabaasa afaaniin ykn barreeffamaan hirmaattoota qorannicha waliin walqabsiisuu danda'u keessatti eeruun hin kennamu.

**7. Mirgoota:** Qorannoon kanaaf hirmaannaan guutummaatti fedhii ofiitiin kan raawwatamu dha. Qo'annoo kana irratti hirmaachuu fi dhiisuu kee labsuuf mirga qabda. Yoo hirmaachuuf murteessite yeroo barbaaddetti qorannicha keessaa ba'uuf mirga qabda kunis faayidaa kasaaraa karaa biraatiin siif malu kamiyyuu si hin mallatu. Gaaffii deebii kennuu hin barbaanne kamiyyuu deebisuun si hin barbaachisu.

**8. Teessoo quunnamtii:** Yeroo kamiyyuu waa'ee qorannichaa ykn hojimaata gaaffiin ykn gaaffii yoo jiraate na qunnamaa: E- mail: sisaygaramu83@gmail.com, Lakkoofsa Mobayilaa 0940770023/0921316710, Koree Gamaaggama Naamusa Qorannoo Fayyaa Dhaabbilee Teessoo Waajjira Bilbila: 0254662011 P.O.BOX: 235, Harar, Ethiopia.

**9. Labsii hayyama tola ooltummaa qabu:** Waraqaa odeeffannoo hirmaattotaa dubbiseera/ naaf dubbifameera. Kaayyoo qorannichaa, hojimaata, balaa fi faayidaa, dhimmoota iccitii, mirga hirmaachuu fi teessoo quunnamtii gaaffii kamiifuu sirriitti hubadheera. Wantoota ifa hin taane ta'uu danda'aniif gaaffii akkan gaafadhu carraan naaf kennameera. Yeroo barbaadetti qo'annoo keessaa ba'uuf ykn gaaffii ani hin barbaanne kamiyyuu deebisuuf mirga akkan qabu naaf himameera. Kanaaf, qorannoo kana irratti hirmaachuuf fedhii kootiin hayyama koo qubee jalqabaa (mallattoo) kootiin nan ibsa.

Maqaa fi mallattoo hirmaataa: _____ _____ Guyyaa _____

Maqaa fi mallattoo Walitti qabaa Odeeffannoo: _____ _____Guyyaa _____

**Tumsa keessaniif Galatooma**

**8.7. Afaan Oromo version participant information sheet and informed voluntary assent obtained from parents form for a minor (age < 18 years)/vulnerable individual to be signed by his/her legal competent representative (e.g.: -parent/guardian)**

**1. Seensa:** Maqaan koo----------.Qorannoo Hospitaala yuuniversitii Ispeeshaalaayizdii waliigalaa Hiwot Faanaa keessatti gaggeeffamaa jiruuf Sisay Geremew Gurmuu Yunivarsiitii Haramayaa, Kolleejjii Fayyaa fi Saayinsii Fayyaatti digirii lammaffaa barachaa jiruuf ragaa walitti qabaa ta'ee hojjechaa jira. Mucaan keessan qorannoo kana irratti hirmaataa akka ta'uuf akka tasaa filatamerraa. Waa'ee qorannichaa fi hirmaannaa daa'imaa akka isiniif ibsuuf xiyyeeffannoo keessan akka naaf kennitan kabajaan isin gaafadha.

**2. Mata duree qorannichaa:** Tatamsa'ina Saalmooneelaa fi Shigeelaa, sababoota walqabataniifi haala farra maaykiroobiyaanii dhukkubsattoota HIV Hospitaala Yuunivarsiitii Ispeeshaalaayizdii waliigalaa Hiwot Fana, Harar, Ethiopia keessatti yaalamaa jiran adda baasuuf.

**3. Kaayyoo qorannichaa:** Argannoon qorannoo kanaa dhukkubsattoota HIV dhukkuba Salmonella fi Shigella qaban Hospitaala Yuunivarsiitii Ispeeshaalaayizdii waliigalaa Hiwot Fana keessatti wal'aanuuf barbaachisummaa guddaa qabaata. Kana malees, kaayyoon qorannoo kanaa qorataa ijoo sagantaa maaykiroobaayoloojiitiif akka barbaachisummaa gartokkeetti barruu qorannoo barreessuu dha.

**4. Adeemsaa fi yeroo:** Odeeffannoo barbaachisaa qorannichaaf gargaaru naaf kennuu dhaaf gaaffilee fayyadamuun daa'ima keessan af-gaaffii nan godha. Gaaffilee 18 gaafadhee bakka ani gaaffii fi deebii gochuun guutu deebisuuf nan gaafadha. Af-gaaffiin kun gara daqiiqaa 18 kan fudhatu ta'a. Saamuda sagaraa dhuma gaaffilee irratti barbaachisaa dha. Kanaaf yeroo kana na qusachuun adeemsa kana mucaa keessan irratti akkan raawwadhu akka naaf hayyamtan kabajaan isin gaafadha.

**5. Balaa fi faayidaa:** Miidhaan qorannoo kana irratti hirmaachun daa'ima keessan irraa gahuu baay'ee xiqqaa dha yookaan hin jiru; garuu yeroo kee irraa daqiiqaa muraasa qofa fudhata. Qorannoon kun hirmaachuuf kaffaltiin kallattiin hin jiraatu. Garuu argannoon qorannoo kanarraa argame Hospitaalli Hiwot Fana dhukkubsataan HIV yeroo gosa baakteeriyaa salmonella ykn shigellaan qabaman yaala qoricha barbaachisaa ta'een akka yaaluuf gargaaruu danda'a.

**6. Iccitii:** Odeeffannoon qorannoo kana irraa walitti qabnu iccitii ta'a. Odeeffannoon daa'ima keessan ykn ofuma keessan addatti adda baasu hin jiraatu. Argannoon qorannichaa hawaasa qorannichaaf waliigalaa kan ta'u yoo ta'u, namoota dhuunfaa ykn mana jireenyaa adda ta'e kan hin calaqqisiifne ta'a. Gabaasa afaaniin ykn barreeffamaan hirmaattoota qorannicha waliin walqabsiisuu danda'u keessatti eeruun hin kennamu.

**7. Mirgoota:** Qorannoo kanaaf hirmaannaan guutummaatti fedhii ofiitiin kan raawwatamu dha. Mucaan keessan qorannoo kana keessatti akka hirmaatu hayyamuu fi dhiisuu isaa labsuuf mirga qabdu. Yoo daa'ima kee qorannoo kanaaf hayyamte, yeroo barbaaddetti qo'annoo keessaa baasuuf mirga qabda kunis faayidaa ati/mucaan kee karaa biraatiin mirga qabdu kamiyyuu si/mucaa kee irraa hin hambisuu.

**8. Teessoo quunnamtii:** Waa'ee qorannichaa ykn hojimaata yeroo kamiyyuu gaaffiin ykn gaaffii yoo jiraate: Teessoo qorataa: Maqaa: Sisay Geremew Gurmu, Lakkoofsa moobaayilaa 0940770023/0921316710, Teessoo E-mail: Sisaygaramu83@gmail.com. Teessoo Koree Gamaaggama Naamusa Qorannoo Fayyaa Dhaabbilee (IHRERC) bilbila Waajjira: 0254662011, P.O.BOX: 235, Harar, Ethiopia.

**9. Labsii hayyama tola ooltummaa beekumsa qabu:**

Waraqaa odeeffannoo hirmaattotaa dubbiseera/ naaf dubbifameera. Kaayyoo qorannichaa, hojimaata, balaa fi faayidaa, dhimmoota iccitii, mirga hirmaachuu fi teessoo quunnamtii gaaffii kamiifuu sirriitti hubadheera. Wantoota ifa hin taane ta'uu danda'aniif gaaffii akkan gaafadhu carraan naaf kennameera. Yeroo barbaadetti mucaa koo qo'annoo keessaa baasuuf ykn gaaffii ani hin barbaanne kamiyyuu deebisuuf mirga akkan qabu naaf himameera. Kanaafuu, mucaan koo

qorannoo kana irratti akka hirmaatu (hirmaatu) hayyama fedhii kootiin qubee jalqabaa (mallattoo) kootiin nan labsa.

Maqaa fi mallattoo warraa/guddistuu: _____ _____ Guyyaa: _____.

Maqaa fi mallattoo Walitti qabaa Odeeffannoo: _____ _____ Guyyaa: _____

**Tumsa keessaniif Galatooma**

**8.8. English version Questionnaire for Data Collection**

Identification or code number _____

Name of Health Facility HFCSUH

Name of data collector _____

Date of data collection_____

CD4 Count----------

WHO clinical stage----------

Adherence to drug treatment----------

Questionnaire format to be filled for research purposes by HIV patients attending ART treatment at Hiwot Fana Comprehensive Specialized Hospital only.

| | **Part I: Identification of Socio-demographic Information** | | |
|---|---|---|---|
| **Code** | **Questions** | **Response** | **Remark** |
| 101 | Age in years | _____ | |
| 102 | Sex | 1. Male<br>2. Female | |
| 103 | Place of residence? | 1. Urban<br>2. Rural | |
| 104 | What is your marital status? | 1. Single<br>2. Married<br>3. Divorced<br>4. Widowed | |

| | | | |
|---|---|---|---|
| 105 | What is your Ethnicity? | 1. Oromo<br>2. Amhara<br>3. Gurage<br>4. Harari<br>5. Others specify---------- | |
| 106 | What religion you are following? | 1. Muslim<br>2. Orthodox<br>3. Protestant<br>4. Other(specify)---------- | |
| 107 | What is your Educational status? | 1. Cannot read and write<br>2. 1-8 Grade<br>3. 9-12 Grade<br>4. College and above | |
| 108 | What is your Occupational status? | 1. Gov't employee<br>2. housewife<br>3. Merchant<br>4. Student<br>5. Farmer<br>6. Other(specify)---------- | |
| 109 | What is your Family size? | _____ | |

| Code | Part 2:Factor associated with *Salmonella* and *Shigella* infection | | |
|---|---|---|---|
| 110 | What is your source of drinking water? | 1. Domestic well water<br>2. River water<br>3. Public hand pump water<br>4. Tap water | |
| 111 | Do you have Latrine (private)? | 1. Yes<br>2. No | If yes |
| 112 | Do you wash your hands after the latrine? | 1. Yes<br>2. No | |
| 2113 | Do you wash your hand before meals? | 1. Yes<br>2. No | |
| 114 | Do you eat uncooked food? | 1. YES<br>2. NO | |
| 115 | Do you have a habit of drinking raw milk? | 1. YES<br>2. NO | |
| 116 | Do you have a habit of eating raw meat? | 1. YES<br>2. NO | |
| 117 | Do you have a habit of eating raw vegetables and fruits? | 1. YES<br>2. NO | |
| 118 | Do you have domestic animals in the house? | 1. YES<br>2. NO | |

**8.9. Amharic version questionnaire for Data Collection**

መለያ ኮድ ቁጥር _____

የጤና ተቋም ስም HFCSUH

የመረጃ ሰብሳቢው ስም _____

መረጃ የተሰበሰበበት ቀን_____

CD4 Count----------

WHO clinical stage----------

Adherence to drug treatment----------

በሂወት ፋና ስፔሻላይዝድ ሆስፒታል ብቻ በሚታከሙ የኤችአይቪ ታማሚዎች ለምርምር ዓላማ የሚሞሉ መጠይቅ ፎርማት።

| | **ክፍል አንድ : ማህበረ ህዝብ ባህሪያት** | | |
|---|---|---|---|
| **ኮድ** | **ጥያቄዎች** | **ምላሽ** | **አስተውል** |
| 101 | ዕድሜ በዓመታት | _____ | |
| 102 | ፆታ | 1. ወንድ<br>2. ሴት | |
| 103 | የ መኖሪያ ቦታ ? | 1. ከተማ<br>2. ገጠር | |
| 104 | የጋብቻ ሁኔታዎ ምንድነው? | 1. ያላገባ<br>2. ያገባ<br>3. የተፋታ<br>4. ባል የሞተባት | |

| | | | |
|---|---|---|---|
| 105 | ብሄር ምንድን ነው? | 1. ኦሮሞ<br>2. አማራ<br>3. ጉራጌ<br>4. ሐረሪ<br>5. ሌላ (ይግለጹ)---------- | |
| 106 | የትኛውን ሃይማኖት ነው የምትከተለው? | 1. ሙስሊም<br>2. ኦርቶዶክስ<br>3. ፕሮቴስታንት<br>4. ሌላ(ይግለጹ)---------- | |
| 107 | የትምህርት ደረጃዎ ምን ያህል ነው? | 1. ማንበብ እና መጻፍ አይችሉም<br>2. 1-8 ክፍል<br>3. 9-12 ክፍል<br>4. ኮሌጅ እና ከዚያ በላይ | |
| 108 | የእርስዎ የሙያ ደረጃ ምን ያህል ነው? | 1. የመንግስት ሰራተኛ<br>2. የቤት እመቤት<br>3. ነጋዴ<br>4. ተማሪ<br>5. አርሶ አደር<br>6. ሌላ (ይግለጹ)--------- | |
| 109 | የቤተሰብዎ መጠን ስንት ነው? | _____ | |

| | | | |
|---|---|---|---|
| | **ክፍል 2: ከሳልሞኔላ እና ከሺግላ ኢንፌክሽን ጋር የተዛመደ ምክንያት** | | |
| 110 | የመጠጥ ውሃ ምንጭዎ ምንድነው? | 1. የቤት ውስጥ ጉድጓድ ውሃ<br>2. የወንዝ ውሃ<br>3. የህዝብ እጅ ፓምፕ ውሃ | |

| | | | |
|---|---|---|---|
| | | 4. የቧንቧ ውሃ | |
| 111 | መጸዳጃ ቤት አለህ (የግል)? | 1. አዎ<br>2. አይደለም | እሺ ከሆነ |
| 112 | ከመጸዳጃ(ሽንት) ቤት በኋላ እጅዎን ይታጠባሉ? | 1. አዎ<br>2. አይደለም | |
| 113 | ከምግብ በፊት እጅዎን ይታጠባሉ? | 1. አዎ<br>2. አይደለም | |
| 114 | ያልበሰለ ምግብየመመገብ ልማድ አለህ? | 1. አዎ<br>2. አይደለም | |
| 115 | ያልፈላ ወተት የመጠጣት ልማድ አለህ? | 1. አዎ<br>2. አይደለም | |
| 116 | ጥሬ ሥጋ የመብላት ልማድ አለህ? | 1. አዎ<br>2. አይደለም | |
| 117 | ጥሬ አትክልቶችን እና ፍራፍሬዎችን የመመገብ ልማድ አለህ? | 1. አዎ<br>2. አይደለም | |
| 118 | ቤት ውስጥ የቤት እንስሳ አለህ? | 1. አዎ<br>2. አይደለም | |

**ስለ ምላሽዎ እናመሰናለን**

**8.10. Afaan Oromo Language Questionnaire for Data Collection**

Lakkoofsa koodii eenyummaa _____.

Maqaa Dhaabbata Fayyaa HFCSUH

Maqaa nama odeeffannoo walitti qabuu _____.

Guyyaa odeeffannoon itti walitti qabame_____.

CD4 Count----------

WHO clinical stage----------

Adherence to drug treatment----------

Gaaffilee dhukkubsattootni HIV Hospitaala yuuniversitii Ispeeshaalaayizdii waliigalaa Hiwot Faanaa qofatti yaalamaniin qorannoof akka guutamuf qophaa'ee.

| | **Kutaa 1ffaa- haala jireenyaa fi Hawwaasummaa** | | |
|---|---|---|---|
| **Koodii** | **Gaaffilee** | **Deebii** | **Mirkanef annaa** |
| 101 | Umuriin waggaa dhaan | _____ | |
| 102 | Salaa | 1 Dhiira<br>2 dhalaa | |
| 103 | Bakka dhalootaa? | 1. Magaalaa<br>2. Baadiyyaa | |
| 104 | Haalli gaa'ela keessanii maali? | 1) Qophaa'e (Kan hin fuune )<br>2) Fuudhaa fi heeruma (Kan fuudhe) | |

| | | | |
|---|---|---|---|
| | | 3) Hiikkaan (Kan hike )<br>4) haadha manaa dhirsi irra du'e | |
| 105 | Sabummaan keessan maali? | 1. Oromoo<br>2. Amaaraa<br>3. Gurage<br>4. Hararii<br>5. kaan biroo (ibsu) ---------- | |
| 106 | Amantaa kam hordofaa jirta? | 1. Muslima<br>2. Ortodoksii<br>3. Pirootestaantii<br>4. Kan biroo(ibsu) ---------- | |
| 107 | Haalli Barnootaa keessan maali? | 1. Dubbisuu fi barreessuu kan hin dandeenye<br>2. Kutaa 1-8<br>3. Kutaa 9-12<br>4. Kollejii fi isaa ol | |
| 108 | Hojiin yookaan dalagaan keessan maali? | 1. Hojjetaa mootummaa<br>2. haadha manaa<br>3. Daldalaa<br>4. Barataa<br>5. Qonnaan bulaa<br>6. Kan biraa (ibsu) ---------- | |
| 109 | Baay'ini maati kessani meqaa? | _____ | |

| **Kutaa 2ffaa: Qabxii infekshinii saalmooneelaa fi shigeelaa wajjin walqabatu** |
|---|

| | | | |
|---|---|---|---|
| 110 | Maddi bishaan dhugaatii keessanii maali? | 1. Bishaan boolla bishaanii mana keessaa<br>2. Bishaan lagaa<br>3. Bishaan harkaan paampii ummataa<br>4. Bishaan tubaa | |
| 111 | Mana fincaanii ni qabduu (kaan dhunfaa keessanii)? | 1. Eeyyee<br>2. Lakki | Yo eeyyee |
| 112 | Mana fincaanii booda Harka keessan dhiqattu? | 1. Eeyyee<br>2. Lakki | |
| 113 | Nyaata nyaachuun dura harka kee dhiqattaa? | 1. Eeyyee<br>2. Lakki | |
| 114 | Nyataa hin bilchatin nyatani bekuu? | 1. Eeyyee<br>2. Lakki | |
| 115 | Anaan hin dafiin ni dhugdu? | 1. Eyyee<br>2. Lakki | |
| 116 | Foon dheedii (hin bilchatin) ni nyatu? | 1. Eyyee<br>2. Lakki | |
| 117 | Kuduraalee fi muduraalee hin bilchatin ni nyatuu? | 1. Eyyee<br>2. Lakki | |
| 118 | Manaa kessan horiin ni jiraa? | 1. Eyyee<br>2. Lakki<br>**Galatooma waan naaf debistanif** | |

### 8.11. Laboratory Standard Operating Procedures (SOP) for Stool sample collection and transferring to transport medium.

**A. Stool sample collection Procedure:** 1. Instruction was given on how to collect samples for study subjects to make sure no urine, water, soil, or other material gets in the container, and to wash the hands before sample collection. A stool sample was taken at any time of collection. 2. Label the container with the date, name, and number of the patient and the time of collection. Give the patient a sterile, dry, wide-necked, tight-fitting, leak-proof container. Request a 2-gram of stool sample or 2 ml for a liquid stool sample. Deliver the specimen with a request form to the laboratory. When immediate delivery to the laboratory is not possible.

**B. Procedure for transferring to transport medium:** 1. Chill the tube of Cary Blair transport medium by placing it on ice packs or in the refrigerator for 1 - 2 hours before collecting the specimen. Note: Remove the wrapper from the handle end of the sterile swab. Do not touch the tip of the swab. 2. Collect a small amount of stool by inserting a sterile cotton or polyester-tipped swab into the stool and rotating it. Immediately insert the swab into the transport medium. The swab should be pushed completely to the bottom of the tube of the transport medium. 3. Break off the top portion of the stick that was in contact with the gloved Fingers. 4. Adhere the specimen label to the container or write it on adhesive tape and secure it to the tube.5. Safely dispose of all contaminated materials. Do not reuse.

**C. Procedure for preparation of carry blarry transport medium:** 1. Suspend 12.6 grams in 991 ml of distilled water.2. Heat to boiling to dissolve the medium completely.3. Cool to 50°C adjust pH to 8.4 if necessary2.Dispense the medium in 7ml amounts in screw-cap bottles of 9 ml capacity's 4. Sterilize by steaming with caps loosened (do not autoclave) at 100°C for 15 minutes. 5. When cool, tighten the bottle caps. Label the bottles.5.Date the medium and give it a batch number. Record the expiry date (6 months from preparation) on each bottle. 6. Store in a cool dark place with the bottle tops screwed tightly.

### 8.12. Culture media preparation Procedure, Result, and Identification

**A. Procedure for XLD Agar Preparation:** 1. Suspend 56.68 grams of the dehydrated medium in 1000 ml of purified or distilled water.2.Heat with frequent agitation until the medium boils. Note: Do Not Autoclave.3.Transfer immediately to a water bath at 50°C.4.After cooling, pour into sterile Petri plates. 5. Culture procedure on XLD 6. The sample was taken by swab 7. Cultured on XLD

**Result and Interpretation:** Typical colonial morphology on XLD Agar is as follows: *Salmonella Typhi* – Red Colonies with Black Centers, *Salmonella choleraesuis* – Red Colonies**,** *Shigella sonnei* – Red Colonies, *Shigella flexneri* – Red Colonies. *Shigella boydii* Red – Colonies shigella dysentery – Red Colonies

**B. Procedure for Preparation Selenite F Broth:** 1. Add 4.0 gm sodium selenite powder to distilled/deionized water. 2. Add the remaining 19.0 gm powder to the above solution and bring volume to 1.0 liter.3.Gently heat and bring to boiling.4.Dispense into sterile test tubes, at least of a depth of 5 cm.5.Sterilize in boiling water bath or at 0-psi pressure at 100°C for 10 minutes. Do Not Autoclave.6.Cool to room temperature before use.

**Procedure for Inoculation of Selenite F Broth:-**1. Inoculate the tube of selenite with 1-2 gm of stool sample and emulsify the broth.2. Incubate in ambient air at 35-37°C for up to 24 hours.

**Result Interpretation:** Selenite F Broth After incubation, record the growth of organisms, indicated by turbidity in the medium. Positive Result: Colorless, Good Growth. Examples: *Salmonella typhimurium, Shigella sonnei, and Salmonella enteritidis*. Negative Result: pink with bile precipitate, Inhibited or no growth.

**Quality Control on Selenite F Broth:** Positive control: *Salmonella typhimurium* ATCC® 14028. Good Growth. Negative control: *Escherichia coli* ATCC® 25922. Inhibited or no growth.

**C. Preparation procedure for MacConkey Agar:** 1. Suspend dehydrated powder in water (53.53 grams in 1000 ml of distilled water). 2. The medium is boiled for a few seconds until the ingredients are completely dissolved.3. Sterilize by autoclaving at 15 lbs (121 ° C) pressure for 15 minutes.4. Cool to 47 ° C, and mix well before pouring into sterile Petri dishes.

**Result Interpretation:** Lactose fermenters, colonies- Red or Pink, Non-fermenters - No color change, *Salmonella*, and *Shigella* - Non-fermenters.

**D. Procedure for preparation of Trypticase Soy Agar (TSA):** 1. Suspend dehydrated powder, in water (40 grams in 1000 ml of purified/distilled water). 2. The medium is boiled for a few seconds until the ingredients are completely dissolved. 3. Finally sterilize by autoclaving at 121℃ for 15 minutes.4. Cool to 45-50℃ and dispense aseptically into sterile Petri dishes. Date the medium and assign it a lot number.

**8.13. Laboratory procedure for biochemical testing**

**A. Procedure for preparation of Tryptophan Broth and procedure for Indole test**

**Procedure for preparation of Tryptophan Broth: -** 1. Dissolve 16g Tryptophan Broth in one-liter water by heating 2. Dispense 3 ml per test tube. Close the tubes with cotton plugs, plastic or metal caps 3. Autoclave for 15 min at 121 +/- 3℃.

**Procedure for Indole Test: -** 1. Take sterilized test tubes containing 4 ml of tryptophan broth.2.Inoculate the tube aseptically by taking the growth from 18 to 24 hrs. 3. Incubate the tube at 37℃ for 24-28 hours.4.Add 0.5 ml of Kovac's reagent to the broth culture.5.Observe for the presence or absence of a ring.

**Result and Interpretation: Positive: -**pink to red color (cherry-red ring) in the reagent layer on top of the media, **Negative**: -No color change, *Salmonella* species - Negative, *Shigella* species- Variable.

**B. Procedure for preparation of MIO (Motility, Indole, and Ornithine) Medium and procedure for MIO test.**

**Procedure for preparation of MIO (Motility, Indole, and Ornithine) Medium: -** Suspend 31.02 grams in 1000 ml distilled water. Heat to boiling to dissolve the medium completely.

Dispense in test tubes in 5 ml amounts. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Cool the tubes in an upright position.

**Procedure for MIO test.:-** Inoculate the tube aseptically by taking the growth from 18 to 24 hrs. 3. Incubate the tube at 37°C for 24-28 hours by stab.

**QUALITY CONTROL**

**Positive control**: *Escherichia coli* ATCC® 25922 Growth; Motility: positive, Indole: positive (turns pink after adding Kovacs Reagent), Ornithine: positive (purple throughout tube)

**Negative control:** *Klebsiella pneumoniae* ATCC® 13883 Growth, Motility: negative, Indole: negative, Ornithine: negative (purple top layer, rest of tube yellow)

**C. Procedure for Preparation of Simmons Citrate Agar and Procedure of Citrate Test**

**Procedure for Preparation of Simmons Citrate Agar:** 1. Suspend 24.28 grams in 1000 ml distilled water. 2. Heat, to boiling, to dissolve the medium completely.3.Mix well and distribute in tubes or flasks. For tubes, dispense 4.0 to 5.0 ml into 16-mm tubes.4.Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes.5.Cool in a slanted position (long slant, shallow butt).6.Tubes should be stored in a refrigerator to ensure a shelf life of 6 to 8 weeks.

**The procedure of the Citrate Utilization Test:** 1. Streak the slant back and forth with a light inoculum picked from the center of a well-isolated colony.2. Incubate aerobically at 35 to $37^0$C for 18-24 hours.3. Observe a color change from green to blue along the slant.

**Result Interpretation:** Positive test-The color of the medium changes from green to bright blue. Negative test- The color of the medium remains unchanged (green)**,** *Shigella species*- Green**,** *Salmonella species*-Blue.

**D. Procedure for preparation of Urea Agar and Procedure for Urease Test**

**Procedure for preparation of Urea Agar:** 1. Suspend 24 grams in 950ml of distilled water 2. Heat until completely dissolved 3.Autoclave at 121 $c^o$ for 15min 4.cool to 50$c^o$. 5. Aseptically add 50ml of urea 40% supplement. Forty percentage 40% = 40gram urea powder with 100ml water.

**The procedure of the Urease test (Christensen's (modified) urea Agar Base:** 1. Streak the entire slant surface with a heavy inoculum from an 18-24 hour pure culture. Do not stab the butt, as it will serve as a color control.2. Incubate tubes with loosened caps at 35°C.3. Observe the slant for a color change at 6 hours and 24 hours unless specified for longer incubation.

**Result Interpretation:** A Positive result -bright pink color (fuchsia), Negative result- no color changes

**Quality Control of Urease Test:** Positive: *Proteus vulgaris* (ATCC13315), Negative: *Escherichia coli* (ATCC25922)

**F. Procedure for Preparation of Triple Sugar Iron agar and Procedure for TSI test**

**Procedure for Preparation of Triple Sugar Iron Agar*:*** 1. Suspend 65 g of the powder in 1 L of purified water. Mix thoroughly.2. Heat with frequent agitation and boil for 1 minute to completely dissolve the powder.3. Dispense into tubes and autoclave at 121°C for 15 minutes. 4. Cool in a slanted position so that deep butts is formed. 5. Test samples of the finished product for performance using stable, typical control cultures.

**Procedure for TSI test:** 1. Obtain a slant of TSIA.2. Using an inoculating needle, stab your assigned organism into the butt of the TSIA slant. As you remove the inoculating needle, drag it in a zigzag pattern up the surface of the slant portion of the tube.3. Incubate the slant for 24-48 hours. After the incubation period, record any changes in the tube.4. Interpretation.

**Results from Carbohydrate Fermentation:** Slant color/butt color -Slant color indicates the fermentation of lactose and/or sucrose. Butt color indicates -the fermentation of glucose, the Production of gas - Agar shows bubbles or may split, Production of H2S-a blackening of the medium.

**F. Procedure for Preparation of LIA and Lysine decarboxylase test procedure**

**Preparation of LIA medium:** 1. Take a suspension of 14.02 grams in 1000 ml of distilled water.2. If necessary, heat to melt your medium to the point of complete dissolution.3. Inject 5 ml into test tubes with screw caps.4. Sterilize using autoclaving at 15lbs tension (121degC) during 15 minutes.

**The procedure of the Lysine decarboxylase test:** 1. Inoculation of Medium Choose the lysine decarboxylase broth medium.2. Incubate at 35-37 C for 24 - 48-hour.

**Result Interpretation:** Positive reaction- purple colored, Negative reaction- yellow-colored *Salmonella arizonae*- positive reaction, purple color, *Salmonella typhi A*- positive reaction, purple color, *Salmonella paratyphi*- negative reaction, yellow color, *Shigella dysenteriae*-negative reaction, yellow color.

**8.14. Antimicrobial susceptibility testing**

**A. Procedure for Preparation of Mueller Hinton Agar:** 1. Dissolve dehydrated powder, in water (38 grams in 1000 ml of purified / distilled water). 2. The medium is boiled for a few seconds until the ingredients are completely dissolved.3. Sterilize by autoclaving at 15 lbs (121 ° C) pressure for 15 minutes.4. Cool to 47 ° C, and mix well before pouring into sterile Petri dishes.

**B. Standard procedure for drug sensitivity test:** 1. Prepare a saline suspension of the isolate from an overnight incubated agar plate to obtain (0.5) McFarland turbidity (1.5 x 108 cfu/ ml of E. coli ATCC® 25922). 2. Muller Hinton Agar plate should be inoculated within 15 minutes after the inoculums have been adjusted. 3. A sterile cotton swab is dipped into the suspension, rotated several times, and gently pressed onto the inside wall of the tube above the fluid level to remove excess inoculums from the swab. 4. The swab will then be streaked over the entire surface of the agar plate three times, with the plate rotated approximately 60 cm each time to ensure an even distribution of the inoculums. A final sweep of the swab was made around the agar rim. 5. The lid may be left ajar for 3 to 5 minutes but no longer than 15 minutes to allow any excess surface moisture to be absorbed before the drug-impregnated discs are applied.

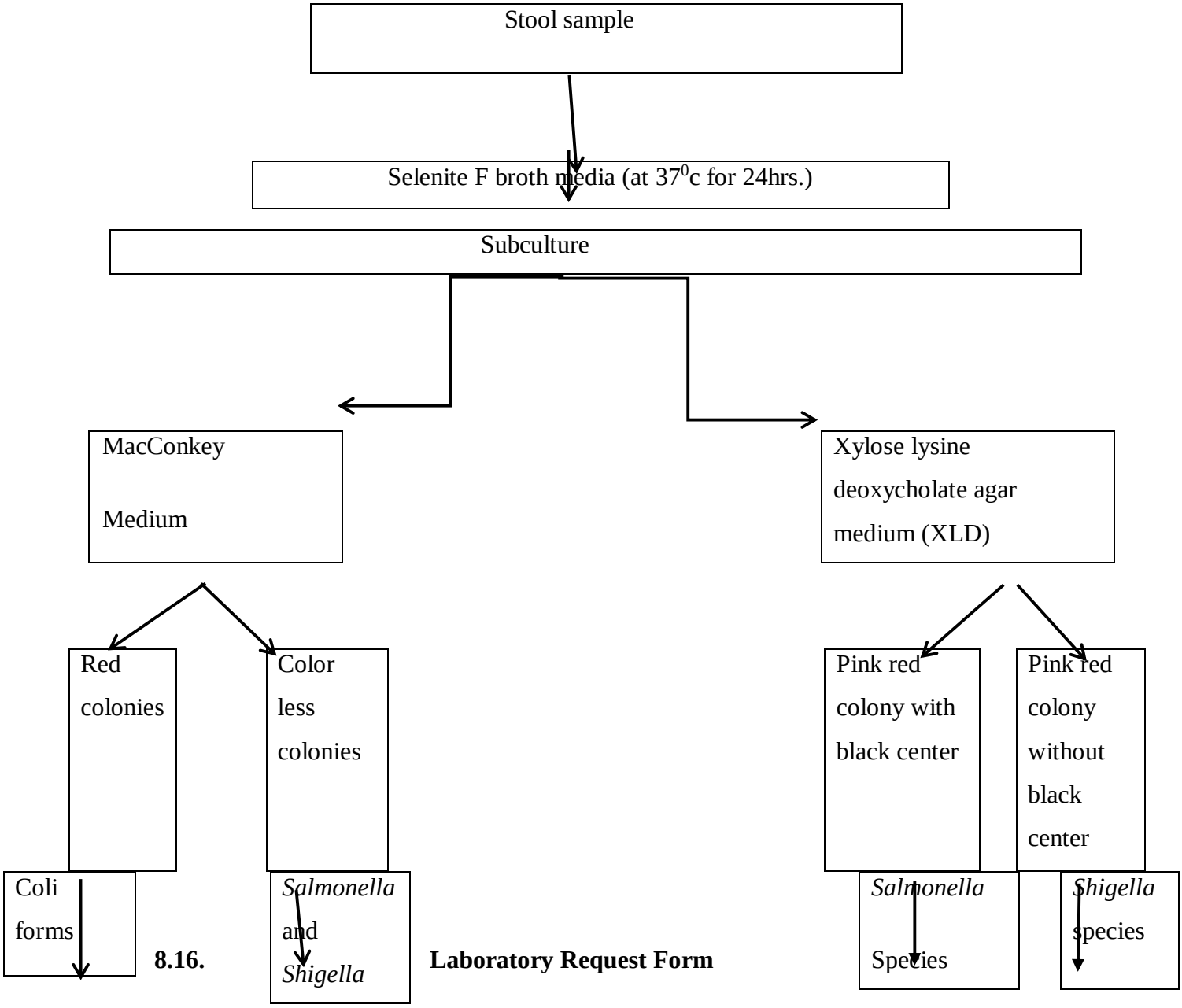
**C. Application of antimicrobial discs to an agar plate:** 1. ideally, this should be done within 15 minutes of the inoculation of plates. 2. The selected antimicrobial discs was dispensed evenly onto the agar plate with the help of a forceps/sterile needle/surgical blade. Flame the tips of the applicator intermittently. Each disc must be pressed down to ensure complete contact with the agar surface.

**D. Result and Interpretation:** Each plate was examined after overnight incubation (16-18 hours), for confluent growth and circular zones of inhibition. The diameters of the zones of complete inhibition was measured to the nearest whole millimeter with ruler. With unsupplemented Muller Hinton Agar (MHA), the measuring device is held on the back of the inverted petri dish, which is illuminated with reflected light located a few inches above a black, non-reflecting background. The zone margin should be considered the area showing no obvious visible growth detectable with the unaided eye. The faint growth of tiny colonies visible only by lens should be ignored. Zone sizes should be measured from the upper inoculated surface of opaque.

**E. Job aid for reading disk diffusion test**

| **Drugs** | **Sensitive** | **Intermediate** | **Resistant** |
|---|---|---|---|
| Ciprofloxacin (CIP-5 µg) | ≥31 | 21-30 | ≤20 |
| Ceftriaxone(30 µg,) | ≥23 | 20-22 | ≤19 |
| Ampicillin(A-10 µg) | ≥17 | 14-16 | ≤13 |
| Co-trimoxazole(SXT−25µg) | ≥16 | 13-16 | ≤12 |
| Amox-clavulanate20/10 µg | ≥18 | 14-17 | ≤13 |
| Chloramphenicol (C-30 µg) | ≥18 | 13-17 | ≤12 |
| Azithromysin (15 µg) | ≥13 | - | ≤12 *S.typhi* |
| | ≥16 | 11-15 | ≤10 *shigella* |
| Gentamycin (GM-10 µg) | ≥15 | 13-14 | ≤12 |
| Tetracycline (Te-30 µg), | ≥15 | 12-14 | ≤11 |
| Nalidixic acid (NA-30µg) | ≥19 | 14-18 | ≤13 |

**8.15. Procedure for Stool culture**




**8.16. Laboratory Request Form**

Test Request Form – To do research at Hiwot Fana Comprehensive Specialized University Hospital among HIV patients.

**1. Patient details**

Name: Id number---------- Age----------sex □ Male □ Female

Address: ---------- Telephone number----------

**2. Sample details:**

Sample type □ Faeces □ Swab

Date: dd/mm/yyyy --------/----------/----------Time (hh/mm) ----------/----------/----------

**3. Clinical information relevant to the laboratory:**

CD4 Count---------- WHO clinical stage----------

Adherence to drug treatment----------Drug therapy: Last dose: (dd/mm/yyyy) ----------
Time :( hh/mm) ---------- Other relevant clinical information: ----------

**4. Examination requested:**

| | |
|---|---|
| Profile test | Microbiology |
| Examination(s) requested. | □ Microscopy/Culture/Sensitivity |
| Consistency of samples | 1. Watery 2. Mucoid 3. Bloody<br>4. Blood + mucoid |

.

**5. Result of test**

| **Test** | **Result** | |
|---|---|---|
| **Selenite F broth** | □ turbidity | □No turbidity |

| | | | | |
|---|---|---|---|---|
| **XLD culture** | □Positive<br>□ Red with black center<br>□ Red only | | □Negative<br>□No colony | |
| **MacConkey agar** | □ Non lactose fermenter | | □.Lactose fermenter | |
| **Biochemical test** | Indole test | | □ Positive (Red ring) | □ Negative(Colorless ring) |
| | Motility | | □Motile | □Non motile |
| | Ornithine | | □Positive □Purple | □Negative □yellow |
| | Citrate test | | □Positive □Blue | □Negative □ Green |
| | Lysine Iron Agar | Lysine De amylase (LDA) | □Positive □ yellow | □Negative □Purple |
| | | Lysine Decarboxylase test(LDC) | □Positive □Purple | □Negative □ yellow |
| | Urease test | | □Positive □ Pink | □Negative □ Yellow |
| | TSI | □K/A Red slant/ yellow butt | □K/K Red slant /Red butt | □A/A Yellow slant /Yellow |
| | | $H_2S$ | □Positive (Blackening ) | □Negative |
| | | Gas production | □ Crack, bubble, displaced medium | □No crack Negative |

| **Sensitivity test** | **Antibiotic disk** | **Sensitive** | **Intermediate** | **Resistant** |
|---|---|---|---|---|

| | | | | |
|---|---|---|---|---|
| | CIPROFLOXACIN (CIP-5 µg) | □≥31 | □21-30 | □≤20 |
| | CEFTRIAXONE(30 µg,) | □ ≥23 | □20-22 | □≤19 |
| | AMPICILLIN(A-10 µg) | □ ≥17 | □14-16 | □≤13 |
| | CO-TRIMOXAZOLE(SXT−25 µg) | □ ≥16 | □11-15 | □≤10 |
| | AMOX-CLAVULANATE(20/10 µg) | □ ≥18 | □14-17 | □≤13 |
| | CHLORAMPHENICOL (C-30 µg) | □ ≥18 | □13-17 | □≤12 |
| | Azithromysin (15 µg) | □ ≥13 | - | □≤12 S.typhi |
| | | □ ≥16 | □11-15 | □≤10 shigella |
| | GENTAMYCIN (GM-10 µg) | □≥15 | □13-14 | □≤12 |
| | TETRACYCLINE (Te-30 µg), | □≥15 | □12-14 | □≤11 |
| | NALIDIXIC ACID (NA-30 µg), | □≥19 | □14-18 | □≤13 |

| **6. Conformation of *Salmonella* species** | | ***Salmonella* species □** | ***S. typhi* □** | ***S. para typhi A*** | ***S. para typhi B*** | ***S. para typhi C*** |
|---|---|---|---|---|---|---|
| Indole | | □Negative | □Negative | □Negative | □Negative | □Negative |
| Citrate | | □Positive | □Negative | □Negative | □Negative | □Negative |
| LDC test | | □Positive | □Negative | □ Positive | □Positive | □Negative |
| Urease test | | □Negative | □Negative | □Negative | □Negative | □Negative |
| TSI | Slant/butt | □K/A | □K/A | □K/A | □K/A | □K/A |
| | Motility | □Motile | □Motile | □Motile | □Motile | □Motile |
| | $H_2s$ | □Positive | □Positive | □Negative | □Negative | □Negative |
| | Gas | □Positive | □Negative | □Positive | □Positive | □Positive |

| **7. Conformation of *Shigella* species** □ | | | ***Shigella flexneri*** | ***Shigella boydii*** | ***Shigella sonnei*** | ***Shigella dysenteriae*** |
|---|---|---|---|---|---|---|
| Indole | | □Negative | □Variable | □Variable | □Negative | □Variable |
| Motility | | □Non motile | □Non motile | □Non motile | □Non motile | □Non motile |
| Citrate | | □Negative | □Negative | □Negative | □Negative | □Negative |
| Urease test | | □Negative | □Negative | □Negative | □Negative | □Negative |
| *LIA* | LDC | □Negative | □Negative | □Negative | □Negative | □Negative |
| TSI | TSI | □K/A | □K/A | □K/A | □K/A | □K/A |
| | $H_2S$ | □Negative | □Negative | □Negative | □Negative | □Negative |
| | Gas | □No gas | □No gas | □No gas | □No gas | □No gas |

Name of examiner ----------Signature ----------

Date of report examination----------

**8.17. Curriculum Vitae**

## 1. Personal information

Full name SISAY GEREMEW GURMU

Date of birth ----------21/10/1983e.c

Place of birth ----------Lega Dadi

Sex----------Male

Marital status ----------Single

Physical condition ----------Normal

Religion----------Orthodox

Nationality----------Ethiopian

Hobby----------Sport watching, reading

Address---------- Bedeno woreda

Phone number ----------0940770023/0921316710

E-Mail ----------sisaygaramu83@gmail.com

## 2. Educational background

Grade 1-8----------Dale Danbal (from 1990-1998 e.c)

Grade 9-12----------Sendafa Secondary School (from1999 - 2002)

BSC degree----------Wollega University (from 2002-2006 e.c)

**3. Language skill**

Amharic ----------spoken and written ----------excellent

Orominyaa ----------both spoken and written ----------very good

English ----------spoken and written ----------very good

**4. Work experience**

Four years of working experience ----------Medical Laboratory Technologist at Dewele Health Center (from 2007-2010 e.c)

Three years of working experience --------Medical Laboratory Technologist at Bedeno Health Center (from 2011-2013 e.c)

**5. Reference**

Mr Shamsadin tofiq Haramaya University lecturer

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