ADDIS ABABA UNIVERSITY

COLLEGE OF HEALTH SCIENCES

DEPARTMENT OF MEDICAL LABORATORY SCIENCES

Magnitude of bacterial contamination of Ethiopian paper money and coins currency and their antimicrobial susceptibility pattern from food handlers in Nifas Silk Lafto Sub-City, Addis Ababa, Ethiopia

By: Hiko Negeho (BSc)

Advisors:

Mr. Kassu Desta (BSc, MSc, Associate professor, PhD) Signature _____Date_____

Mr. Melese Hailu (BSc, MSc, PhD Fellow, Assistant professor) Signature _____Date_____

A research thesis submitted to the Department of Medical Laboratory Sciences, College of Health Science, Addis Ababa University, in partial fulfillment of Master of Science Degree in Clinical Laboratory Sciences (Diagnostic and Public Health Microbiology).

September, 2021

Addis Ababa, Ethiopia

Addis Ababa University

College of Health Sciences

Department of Medical Laboratory Science

This is to certify that the thesis prepared by **Hiko Negeho**, entitled:

Magnitude of bacterial contamination of Ethiopian paper money and coins currency and their antimicrobial susceptibility pattern from food handlers in Nifas Silk Lafto Sub-City, Addis Ababa, Ethiopia and submitted to the Department of Medical Laboratory Sciences, College of Health Science, Addis Ababa University, in partial fulfillment of Master of Science Degree in Clinical Laboratory Sciences (Diagnostic and Public Health Microbiology) complies with the regulations of the University and meets the accepted standards with respect to originality and quality.

Principal Investigator:

Hiko Negeho (BSc) Signature _____ Date_____

Advisors:

1. Mr. Kassu Desta (BSc, MSc, Associate professor, PhD) Signature _____Date_____
2. Mr. Melese Hailu (BSc, MSc, PhD Fellow, Assistant professor) Signature ____Date____

Examiners:

1. Internal Examiner_____ Signature _____Date _____
2. External Examiner_____ Signature _____Date _____

_____

Chairman of the Department or Graduate Program Coordinator

September, 2021 G.C

Acknowledgement

I would like to thank my advisors **Kassu Desta (BSc, MSc, Associated professor, PhD)** and **Melese Hailu (BSc, MSc, Assistant professor, PhD)** for their guidance, critical reviews and constructive comments. Also, I would like to thank Addis Ababa University and Department of Medical Laboratory Sciences for their support. My sincere gratitude will also extend to Tikur Anbesa Specialty Hospital (TASH) for the primary permission to conduct this research in microbiology laboratory department using the required equipment, supplies and reagents. Also I would like to thank all my data collectors. Finally, my special thanks go to Lema Tafa (BSc, MPH) who supported me throughout my thesis work.

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Abbreviations

AA	Addis Ababa
AAU	Addis Ababa University
AD	Anno Domini
AST	Antimicrobial Susceptibility Test
CFUs	Colony Forming Units
CHS	College of Health Center
ESBL	Extended Spectrum Beta Lactam
CLSI	Clinical and Laboratory Standards Institute
ETB	Ethiopian Birr
IRB	Institutional Review Board
MRSA	Methicillin-resistant Staphylococcus aureus
NSL	Nifas Silk Lafto
PI	Principal Investigator
SC	Sub-City
SPSS	Statistical Package for Social Science
TASH	Tikur Anbesa Specialty Hospital
TSS	Toxic Shock Syndrome
US	United States

Abstract

Background: Money is the items most commonly passed from hand to hand and can contaminate. Simultaneous handling of food and such contaminated currency could result in foodborne infection, and cause of food poisoning. Microorganisms commonly present on banknotes include S. aureus, E. coli, Klebsiella spp., *Streptococcus*, *Acetobacter* spp., *Bacillus* spp., *Salmonella* spp., *Pseudomonas* spp., and *Enterobacter* spp. handling of food and money by meat butchers, Fruit and Juice servers, and waiters, or vendors can have serious consequences as the food they serve is ready to eat.

Objective: To determine the Magnitude of Bacterial contamination of Ethiopian paper notes and coins currency and their antimicrobial susceptibility pattern from food handlers in Nifas Silk Lafto Sub-City, Addis Ababa, Ethiopia, 2020/2021.

Methods: A cross-sectional study was conducted from January 15, 2021 to May 30, 2021 in NSL Sub-city. The sample of currency was randomly collected from butchers, fruit and juice houses, hotels and restaurants. A general of 200 Ethiopian paper cash and coins were collected aseptically into sterile polythene bags and Culturing by inoculated on blood agar, MacConkey agar and chocolate agar. Then isolated bacteria were assessed for Gram reaction, and conducting biochemical tests. Statistical data analysis involved descriptive analysis of bacterial isolates.

Results: A general of 200 Ethiopian paper cash and coins were analyzed for bacterial contamination. 287 different bacteria were isolated from 183 (91.5%) currencies. Types of bacteria isolated from the notes were Coagulase-negative staphylococci (51.60%), Bacillus species (12.20%), S. aureus (8.70%), Entrobacter spp. (8.0%), Citrobactor spp. (3.50%), Klebsella spp. (2.80%), Shigella spp. (2.80%), Serratia (2.40), Streptococcus spp. (1.70), E. coli (1.40%), Salmonella spp. (1.40), Pseudomonas (1.40), Proteus spp. (0.70), and Acitinobacter spp. (0.30). The highest resistance rates in gram-positive bacteria were against penicillin, erythromycin and clindamycin, whereas ampicillin was the most resistance gram-negative bacteria**.**

Conclusion: Ethiopian paper money and coins currency reported that confirmed currency might be a vector playing an important role in the transmission of pathogenic bacteria in the community. Potentially pathogenic bacteria that are highly resistant to the most widely used antibiotics and are a threat to public health.

Key words: Bacterial contaminations, Ethiopia, paper currency, coins currency, antimicrobial susceptibility, food handlers.

1. Introduction

1.1. Back ground

In ancient times, people got the whole thing they needed through bartering. The first money was commodity money before thousands of years ago. Later commodity money was replaced by coins made of silver, copper, gold and bronze and soon after paper money was developed the first time in China **[1, 2].** Globally Money is the most widely used and required service on the world **[1, 3, 4].**

The transfer of paper currency has been the representation of economic exchange since its introduction in China approximately 1000 AD [**1, 5, 6**]. We use money as a medium for exchange of goods and services, settlement of debts, for deferred payments in economic exchanges **[7, 8]**. The Ethiopian cash-money called "Birr" is the second one maximum used cash-money in Africa subsequent to Nigerian "Naira" for items and services **[5, 9]**. The scientists began to theorize that the transmission of money was associated with the transmission of disease, in the late 1800s and early 1900s **[1].** Modern scientific techniques have confirmed these theories and have shown that viable pathogenic bacteria can be isolated on the surfaces of both paper and coin currency **[10, 11]**. Making it a main multiplication medium for various microorganisms and could comprise a major health hazard **[12]**.

Globally, money is one of the items most commonly passed from hand to hand. The general hygiene levels of a community may contribute to the number of microbes found on coins and notes **[13].** Consequently, hand hygiene is considered critical for preventing food outbreaks and healthcare-associated infections **[14]**.

Banknotes and coins are handled by persons of varying health and hygienic standards, and are stored under varying environmental and personal hygienic situations **[14].** Banknotes and coins can also serve as pathogen reservoirs **[7, 15]**. Simultaneous handling of food and money by waiters or vendors can have serious consequences as the food they serve is ready to eat and does not need any further heating and the people eat by ordering that food **[15].** Microorganisms may tolerate on it for longer periods of time **[3, 15].**

The pathogenic bacteria isolated from that cash-money might also additionally reason a extensive type of diseases from meals poisoning, wound and skin infections, droplets during coughing, sneezing, or other materials and placement on dirty surface, poor hand washing post toilet, respiratory and gastrointestinal problems to life threatening diseases such as meningitis and septicemia. In addition, contamination of cash-money also can be traced to dust, soil, water, micro flora of the body of handlers counting machine, atmosphere, storage environment, usage, handling or production (hand, skin, etc.) **[5, 16, 17, 18]**. Furthermore, many people tongue wet their fingers when counting money thus, contaminating their fingers as well as currency notes **[7, 9, 19].** Simultaneous handling of food and such contaminated currency could result in foodborne infection **[5, 9]**. Microbial contamination percent of currency handled by butchers (78.0 %) and food sellers (62.1 %) was also reported from Nepal **[11]**.

Most of the organisms were of Gram-positive rods, cocci and also endospore formers and few Gram-negative rods. Microorganisms commonly present on banknotes include S. aureus, E. coli, Klebsiella sp., a-haemolytic *Streptococcus*, *Acetobacter* sp., *Bacillus* sp., *Salmonella* sp., *Pseudomonas* sp., *Enterobacter* sp., and V. cholera. Some banknotes associated bacteria may cause opportunistic infections while other bacteria are pathogenic and it is also a common cause of food poisoning **[1, 18]**. Staphylococcus epidermidis is usually non-pathogenic, but it is a critical cause of infection in patients who's immunocompromized. Klebsiella pneumoniae is a virulent organism that can cause pneumonia typically along with urinary tract and wound infections, particularly in immunocompromized persons. Also, E. aerogenesa is a pathogenic bacterium that causes opportunistic infections in skin and other tissues **[11]**.

Some gram-negative bacteria can remain as long as eleven days on coins **[11]**. Accumulation of data during the last 20 years shows that pathogens on currency notes could represent a potential cause of food- borne illness **[3, 5]**.

Additionally, market men and women squeeze paper currencies and put them into their dirty pockets. Meat sellers in slaughter houses and at market places usually collect money from buyers with hands contaminated with blood and animal wastes **[20]**. Such money handling habits can bring in microbes to the notes. In most parts of the world, it is believed that the simultaneous handling of food and money contributes to the occurrence of food-related public health incidents **[5]**.

Antimicrobial resistance is a global phenomenon that has resulted in high mortality and morbidity in consequence of treatment failures and increased health care costs **[13]**. While antimicrobial resistance has gradually been increasing, e.g. with Extended- Spectrum Beta

Lactamases (ESBL) producing Escherichia coli and Klebsiella sp. contaminated banknotes and coins contribute to the transmission of these multi- drug resistant microorganisms in the community **[21]**.

Therefore assessing the level of contamination and microbial types in Ethiopian currency notes and coins were develops our knowledge on infection prevention and hand hygiene practice among the wider population who uses currency for our day to day life.

1.2. Statement of the problem

Money is best route for hand-to-hand contamination. Many pathogenic or antibiotic-resistant bacteria have been isolated from various coins and paper money collected from food handlers. In addition, the possibility that terrorists could contaminate banknotes with pathogens and then put those notes back into circulation has been proposed **[19]**. Antimicrobial resistance is a global phenomenon that has resulted in high mortality and morbidity in consequence of treatment failures and increased health care costs **[13]**.

The first problem is that the paper currencies evaluated were 100% contaminated with different microbes including potentially pathogenic Salmonella spp. and S. auras. The presence of high number of pathogenic bacteria could cause food borne diseases like typhoid fever and food poisoning. Isolated pathogenic strains from cash-money were found growing to infective dose within 12-18 hours indicating that paper currencies are among the risk factors to human health. Periodic evaluation of microbial safety of paper currencies is recommended. There is a poor hygienic practice being exercised while handling paper currencies **[5]**.

Additionally, many meals outlets depend closely on money for exchange with high frequency of contact among the currencies and foods **[5]** risking the safety of consumers **[22]**. Though numerous researches mentioned on the level of microbial contamination of money from different parts of the world including Africa, to-date to the authors' knowledge there was no report on its status from Ethiopia **[5].**

Secondly, studies of the contamination of money with microbial agents is lacking in most developing countries. Shortage of information may contribute to the absence of public health policies regarding currency usage, handling, and circulation **[3]**. Thirdly, studies of the contamination of Ethiopian paper notes currencies with microbial agents are very few**.** Fourthly, study of the contamination of Ethiopian coin currencies with microbial agents is not done. Generally, study of the contamination of Ethiopian paper notes and coin currencies with microbial agents is not done in Addis Ababa city. Therefore, the purpose of this study was to assess the bacterial contamination and safety of Ethiopian paper notes and coin currencies collected from meat butchers, Fruit and juice houses, hotels and food restaurants in Addis Ababa City Administration, central Ethiopia.

1.3. Significance of the study

Money is best route for hand to hand contamination during its circulation. Many pathogenic or antibiotic resistant bacteria have been isolated from various coins and paper money collected from food handlers **[19].** Therefore, this study can have important significances to different stakeholders. The major ones include:

- It helps individuals and societies how to use currencies during serving, preparation and handling food to prevent contamination.
- It helps community to avoid transmission of bacterial infection from paper notes and coin currencies.
- It helps the health managers, planners and policy makers to design strategies on hygiene and care of contact money and food handling.
- It helps Banks and business organizations make use of machines made for the purpose of counting money and they should avoid using saliva for wetting the notes.
- Serves as a reference study for other researchers that would like to explore further in a similar area.

2. Literature review

Paper currency is extremely important since it is widely exchanged for goods and services worldwide **[11, 23]**. It is one of the earliest and most important inventions crucial to the development of trade **[24]**. Paper currency and coins can serve as agents for transmission of microorganisms and are frequently and freely passed from person to person **[25]**.

Paper notes and coins are commonly handled and transferred over large geographical areas there is the potential to readily disseminate contamination across the world **[7, 15]**.

Both paper banknotes and coins offer a larger surface area to harbor bacteria and microorganisms, and the hygienic status of currency has been a scourge to some for over a century **[3, 14, 15]**.

Many authors have raised the concern that banknotes and coins could serve as vectors for the transmission of disease-causing microorganisms **[14]**. Microbial contaminants may be transmitted directly; via hand to hand contact, or indirectly, through food or other non-living objects **[14]**.

According studies from around the world have reported shows high rates of microbial contamination of currency notes in circulation **[25, 26]** While every location contained endemic bacterium, the microorganisms most commonly isolated on paper money included members of the family Vibriocholerae, Mycobacterium tuberculosis, Staphylococcus sp., Micrococcus sp., Enterobacteriacea, Corynebacterium sp., and Bacillus sp. **[12]**. As study shows common background contaminants of paper money were environmental organisms such as gram-positive flora (especially Bacillus sp.) and those arising from human normal skin flora such as Staphylococcus aureus **[12]**.

Microbial contamination of paper money is not only limited to developing nations. Several studies from the United States reported contamination of coins and paper bills and revealed the presence of pathogenic microbes like Staphylococcus aureus, Escherichia coli, and Klebsiella enterobacter **[25]**. A study of US currency isolated 93 types of bacteria (belonging to the species Staphylococcus, Streptococcus, Enterobacter, Acinetobacter, Pseudomonas, Bacillus, Diptheroids, Klebsiella *pneumoniae,* and Escherichia *vulneris*) **[12]**.

A study of American coins and currency had shown the presence of pathogenic bacteria on18% of the coins and 7% of the bills **[28].** As the study stated that the cleanest banknotes contained 20 CFUs (colony-forming units) and the dirtiest banknotes contained more than 25,000 CFUs **[27]**. Marketing processes are of particular concern such as food carts, local markets, etc., as operators often organize, serve, and collect money from various consumers without properly washing their hands **[25]**. According to study showed in Nepal in 2009, a significant association has been established between contamination and sources of money from such as minibus drivers, butchers, food sellers, and banks were examined in this study, with the highest degrees of contamination isolated among cash- money from minibus drivers (84.8%), that followed by butchers (78.0%), and food sellers (62.1%). No microbial contamination was found on new currencies obtained from banks **[11]**.

In another study shown in British Columbia Institute of Technology Environmental Health in 2013, Data gathered during the last 20 years show that pathogens on cash-money could represent a potential cause of food borne illness **[25]**. Many food outlets depend on highly on the exchange of paper note for their products. If the same individual is handling both money and food products; especially ready to eat foods, rises the risk of cross-contamination **[22]**. These findings have resulted in a number of changes regarding how food is prepared and handled in the food service production. In some examples, the handling of food and money has been physically separated. In other cases, gloves are used to handle food and bare hands are used to handle the money, or vice versa. In both conditions, employees of food service establishments are often saw handling money and food improperly **[22]**.

Study of microbial contamination of paper currency and coins collected in Kermanshah in 2014 and Bacterial Contamination of paper Currency and Coins in Circulation: A Potential (18). Cash-moey as a potential environmental vehicle for transmitting microorganisms among food related workers in Alexandria, Egypt **[28]**.

According to Bacteria Studies from around the world have reported high rates of microbial contamination of currency notes and coins in circulation **[25, 26].**While every location contained endemic bacterium, the most commonly microorganisms isolated on money included members of the family Enterobacteriaceae, Vibriocholerae, Staphylococcus sp., Mycobacterium tuberculosis, Bacillus sp., Corynebacterium sp. and Micrococcus sp. **[12]**.The common related

Contaminants of currency notes were environmental microbes such as especially Bacillus sp of gram positive flora and like Staphylococcus auras those arising from human normal skin flora **[12]**.

Developing countries take the highest rates of currency contamination. After studies of different researches conducted all over the world, it became clear that poor populations with large, impoverished populations were funding these studies in India, Nepal, Myanmar, Vietnam, and several parts of Africa **[7, 25]**.

A number of studies from the United States reported contamination of coins and paper bills and revealed the presence of pathogenic microbes like Staphylococcus aureus, Escherichia coli, and Klebsiella Enterobacter **[12]**. One such study of US cash-money isolated 93 kinds of bacteria belonging to the species Staphylococcus, Streptococcus, Enterobacter, Acinetobacter, Pseudomonas, Bacillus, Diptheroids, Klebsiella pneumoniae, and Escherichia vulneris **[12]**.

Developing countries have the highest degrees of cash-money contamination. A study conducted in Venda region, South Africa indicated that microbes were isolated from 96% of the used currency collected in the study **[1]**. However, no microbes were isolated from new currencies received directly from the bank. Accordingly, Bacterial contamination of paper note is not only limited to developing countries. The degree to which paper currencies are contaminated is incredible. Studies in different parts of the world have reported high degrees of bacterial contamination of cash-money in transmission. Previous Studies conducted on India rupee **[25]**, Iraqi **[29]**, and Ghanaian **[1]**. Paper notes were contaminated with 100% by potentially pathogenic microbes. 88% to 100%, of the Saudi one Riyal paper currency **[18]**, 75% of Nepal money **[19]**, 96.25% of Palestine money **[30]** and 95% of Nigerian cash counting machines **[1]** were positive for the presence of potentially pathogenic microbes with mixed bacterial growth. The isolation of bacterial agents from paper notes in numerous study confirmed that money might be a vehicle playing main role in the transmission of pathogenic microorganisms in the community.

Various Studies all over the world indicates that paper notes and coins conducted were contaminated with Gram-positive as well as Gram-negative bacteria. As numerous study indicated the culture from the collected paper currency and coins produced various kinds of

bacterial species. For instance, E. *coli*, Proteus sp., S. auras, Pseudomonas sp., Bacillus sp., and Klebsella sp., were isolated from paper notes and coins of India **[25]**, Iraqi **[29]**, Nepal **[11]**, Palestine **[30]**, Saudi Arabia **[3]**, Nigeria **[1, 31]**, Ghana **[4]**, Bangladesh **[16]**. Moreover, Vibrio sp., Salmonella sp., and shigella sp., there were similarly isolated from coins and paper notes **[1, 11, 16, 29, 30, 32]**. The contamination source of the used cashes necessity be from handling and use. Particular concern was the isolation of Shigella and Salmonella from the cash, which indicated fecal contamination. This result supports the theory that individuals who prepare food after handling contaminated currency notes have a higher risk of infecting themselves and others with food borne pathogens **[32]**.

A study conducted in recent in Cameroon reported that there was an important difference in contamination with respect to cash-money denomination, physical state and source. All samples from butchers were contaminated. According to this study the lower denominations showed significantly higher levels of contamination than higher denominations. Similarly dirty banknote was more contaminated than clean cash **[1]**.The studies that conducted in Nigeria have shown that, the Nigerian cash counting machines produced six different microbial species like *Staphylococcus sp., Streptococcus sp., Salmonella typhi, Escherichia coli, Streptococcus pyogenic, Enterococcus sp., Proteus and sp.* ***[1].***

In others study Currencies with different denominations collected from people of various categories that is fish mongers, butchers, roadside vendors, sweepers, carpenters have revealed the presence of different pathogenic bacteria**[1]**.Similarly most of the dirty paper currencies which were collected mainly from the butchers, fish mongers &sweepers were found to be carrying most the pathogenic microorganisms**[1]**. In contrast, Indian currency collected from bank, Municipal Corporation, food sellers, butchers, hospital indicated that currencies used by public (bank, hospital, Municipal Corporation) were found to be extremely contaminated with various pathogenic bacteria followed by the currency used by butchers and food sellers **[25]**.

Another study conducted in Australia shown that the lower the index values of the money, the higher the typical microbial content of the banknote. They more showed that the age of the money and the material that was used to produce the money influence the number of microbial contaminations **[1]**. The lower denomination notes harbor the highest bulk of infectious agents since they are exchanged more than higher denomination currency **[1]**. Additionally, a Study

from Nigeria, reported that Contamination was significantly correlated with the denomination of the notes. The lower denomination currencies were more contaminated than higher denomination currencies **[31]**. This result is supported by the other studies that show higher denomination cash indicated lower contamination **[31, 32]**.

Bacterial Pathogens of Concern

Potentially dangerous bacterial agents that have been isolated on paper currency like: Streptococcus and Staphylococcus that have developed resistance to conventional antibiotics**,** E. coli is usually nonpathogenic, but some strains can cause serious food-poisoning infections**,** Enterobacter cloacae are associated with urinary tract and respiratory tract diseases**,** Staphylococcus epidermidis is usually nonpathogenic but can cause infection in patients whose immune system is compromised**,** K. pneumoniae is a virulent bacterium that can cause pneumonia disease, usually along with urinary tract and wound infections, mainly in immunocompromised individuals, Enterobacteraerogenes is a nosocomial and pathogenic bacterium that causes opportunistic infections in skin and other tissues**,** Salmonella choleraesuis can reason for salmonellosis, serious acute gastroenteritis with sudden onset of headache, abdominal pain, diarrhea, nausea, and sometimes vomiting and S. auras can cause a series of infections, from minor skin infections like pimples, impetigo boils, and abscesses, to life-threatening diseases such as pneumonia, meningitis, osteomyelitis, endocarditis, toxic shock syndrome (TSS), and septicemia **[11]**. Though, other bacteria like, Vibrio cholerae, Micrococcus sp., Mycobacterium tuberculosis, Shigella dysentriae and, Corynebacterium sp. were isolated from cash-money too **[4, 32]**. This may cause a wide variety of diseases from food poisoning, skin and wound infections, gastrointestinal and respiratory problems to life threatening diseases such as meningitis and septicemia **[1]**.

The matter of very importance is that not only children and immunocompromised (including those with HIV, undergoing chemotherapy, or taking other medications that suppress the immune system) but also healthy people are susceptible to serious dangers, due to presence of different and plenty pathogens on paper notes **[1]**.

Antimicrobial Resistance

Antimicrobial resistance is a global phenomenon that has resulted in high morbidity and mortality consequently treatment failures and increased health care costs **[33]**. Resistance pattern

of all pathogenic Gram negative bacilli and Gram positive organisms were only one isolate of S. aureus remained MRSA and All the Gram negative bacilli stood resistant to Ampicillin.11 strains of E. coli and 49 strains of Klebsiella remained resistant to Ampicillin. These two bacteria stood found to be quite sensitive to Amikacin, Gentamicin and Ciprofloxacin. Klebsiella stood more resistant to different cephalosporin groups whereas E.coliremained less resistant. Pseudomonas stood quite sensitive to Amikacin, Gentamicin, Ciprofloxacin and cephalosporins **[21]**.

There are many investigation confirms that antibiotic resistant bacteria contaminate currency notes and might play an important role in the transmission of pathogenic microorganisms in addition to in the spread of drug-resistant organisms. Currency notes in circulation are contaminated with various bacteria of which most are resistant to commonly used antibiotics and consequently represents dangers and public fitness threats to the society and individuals handling cash. Study conducted in Ghana indicated different resistance to commonly use antibiotics with Coagulase-negative staphylococci, E. faecalis and Salmonella sp. having high resistivity of 87.5% whereas B. cereus and P. aureginosa indicated 50% sensitivity. That the isolates were shown 100% resistant to Ampicillin, Cefuroxime and Penicillin **[29]**. Coagulase negative staphylococci (CNS), E. faecalis and S. pneumonia indicated 87.5% resistance. B. cereus stood 50% resistant, 37.7% susceptible and 12.5% intermediate to the antibiotics while S. auras was that 75.0% resistant and 25% susceptibility **[29]**.

According to study carried out in University of Uyo Teaching Hospitals S. Africa, from 32 paper notes (naira) thirteen naira (40.6%) provided isolates of S. auras and following resistant range of between 62.5% and 100% to frequently prescribed antibiotics. The comparable set of isolates revealed three (23.1%) to be susceptible and ten (76.9%) to remain resistant to vancomycin **[1]**. As the study revealed Indian cash, among gram positive sp. S. auras indicated 100% ressistance to penicillin. Methecillinresstant S. auras (MRSA) was found to be 36.4% using cefoxitin disc. Of the gram negative microbes isolate, Acinetobacter sp., citrobacter sp., klebsiella sp. and Pseudomonas aeruginosa confirmed 100% sensitivity to gentamicin. Acinetobacter sp. Showed 100% resistance to piperacillin and 75% of the isolates were that resistant to ampicillin and cefoperazon **[34]**.

Paper notes gathered from meat dealers in marketplace of Tanga city of Tanzania confirmations that 28.125% S. auras isolates were that multidrug resistant. S. auras isolates resistant to

Vancomycin likewise resistant to Methicillin **[35]**. Study from Lusaka, Zambia, A general of 205 cash-money was gathered from restaurants and hotels and the prevalence of S. aureus is 25.85%. All Staphylococcus auras isolates indicated multidrug resistance and 2.92% of Vancomycin resistance. That the isolates resistant to vancomycin were likewise resistant to Methicillin **[36]**.

Many antibiotic-resistant bacteria have been isolated from various coins and paper money collected from food handlers **[37]**.

3. Objective

3.1. General objective

To assess the magnitude of Bacterial contamination of Ethiopian paper money and coins currency and their antimicrobial susceptibility pattern from food handlers in Nifas Silk Lafto Sub-City, Addis Ababa, Ethiopia, 2020/2021.

3.2. Specific objectives

- To assess the Magnitude of Bacterial contamination of Ethiopian paper notes and coins currency from food handlers in Nifas Silk Lafto Sub-City, Addis Ababa, Ethiopia, 2020/2021.
- To determine antimicrobial susceptibility pattern for the contamination of Ethiopian paper notes and coins currency in Nifas Silk Lafto Sub-City, Addis Ababa, Ethiopia, 2020/2021.

4. Hypothesis

Null hypotheses [H0]:

- The magnitude and types of microorganism isolated from Ethiopian paper notes and coins is the same with previous study done in Ethiopia.
- The antimicrobial susceptibility pattern for the contamination of Ethiopian paper notes and coins currency is the same with previous study done in Ethiopia.

5. Methodology

5.1. Study Area

The study was conducted in Nifas Silk Lafto Sub-city under Addis Ababa City Administration. Nifas Silk Lafto Sub-city is one of the ten sub-cities in the city. According to 2019/2020 Nifas Silk Lafto Sub-city woreda base plan report, the total population of the sub-city is 414,505, of which 48% male and 52% are female. The sub-city comprises of 13 woredas. In the sub city there are 244 meat butchers, 167 Fruit and juice houses, 84 hotels and 315 bar and restaurants according to Nifas Silk Lafto Sub-city Food and drug administration office report of 2019/2020.

5.2. Study Design and Period

A cross-sectional study design was conducted from January 15, 2021 to May 30, 2021.

5.3. Population

5.3.1. Source population

All Ethiopian paper money and coins handled by food handlers working in meat butchers, fruit and juice houses, hotels and restaurants in Nifas Silk Lafto Sub-City.

5.3.2. Study sample

All Ethiopian paper money and coins handled by food handlers working at selected meat butchers, fruit and juice houses, hotels and restaurants found in the sub-city during data collection period.

5.4. Inclusion and exclusion criteria

5.4.1. Inclusion criteria

Food handlers who have contact with money such as

- waiters,
- meat butchers and
- Fruit and Juice servers were included in the study at selected meat butchers, Fruit and juice houses, hotels and restaurants.

5.4.2. Exclusion criteria

- Food handlers who were on annual leave.

5.5. Study variables

5.5.1. Dependent variables

- Bacterial contamination status of Ethiopian paper money and coins currency.
- Antibiotic susceptibility pattern.

5.5.2. Independent variables: Factors affecting your main outcome

- Meat butcher, Fruit and juice houses, Hotels and Food restaurants
- Denominators of paper money and coins currency
- Old and new paper money and coins currency
- Types of antibiotics

5.6. Sample size calculation and Sampling method

5.6.1. Sample size calculation

Sample size determination for specific objectives is determined by using single proportion formula with assumption of 95% confidence interval (CI), 5% margin of error, by assuming 10% non-response rateand by taking 86.4% proportion of contamination of paper currency notes and coins **[21]**, then by applying the formula the sample size (n) is obtained as below.

$n = (Z\alpha/2)^2 P(1-P))$, Where, n = Sample size

d^2 $Z\alpha/2$ = Critical value =1.96,

d= margin of error =0.05

$$n = \frac{(1.96)^2(0.864 \times 0.136)}{(0.05)^2} = 181$$ and by adding 10% non-response rate, the minimum sample size for this study was 200.

5.6.2. Sampling Method

According to the Pilot study that is carried out around the study area, the sample of currency was randomly collected from meat seller at butcher, fruit sellers' at juice house, waiters of hotels and food restaurants, in NSL Sub-city. In addition, 20 newly minted paper currencies and coin currencies, obtained from Commercial Bank of Ethiopia, Lebu branch was used as negative control. There are a total of 244 meat butchers, 167 Fruit and juice houses, 84 hotels and 315 food restaurants, in all 13 woredas of NSL Sub-City administration. 25 meat butchers houses, 17 Fruit and juice houses, 9 hotels and 78 food restaurants were picked randomly by allocated proportionally based on their numbers.

The number of participants in each institutions/houses/ were allocated proportionally based on their numbers. The first one number was selected randomly by lottery method. All denominations of paper note and one birr coins currency of Ethiopia (A random of Birr 5, Birr 10, Birr 50, Birr 100 and Birr 200 paper notes and Birr 1 coins currency) were collected from each notes and coins. From each of the meat butchers, fruit and juice sellers, hotels and food restaurants that was considered. A total of 200 paper currency notes and coins were sampled and the samples were collected aseptically by engaging food vendors with questionnaires to drop the paper and coin currency into sterile polythene bags. The sterile polyethylene bags were promptly sealed and individuals are given a replacement note equivalent to the denomination they was deposited in the sampling polyethylene bag(s). The polyethylene bags were immediately transported aseptically to the laboratory of the Department of Microbiology, TASH for microbial analysis.

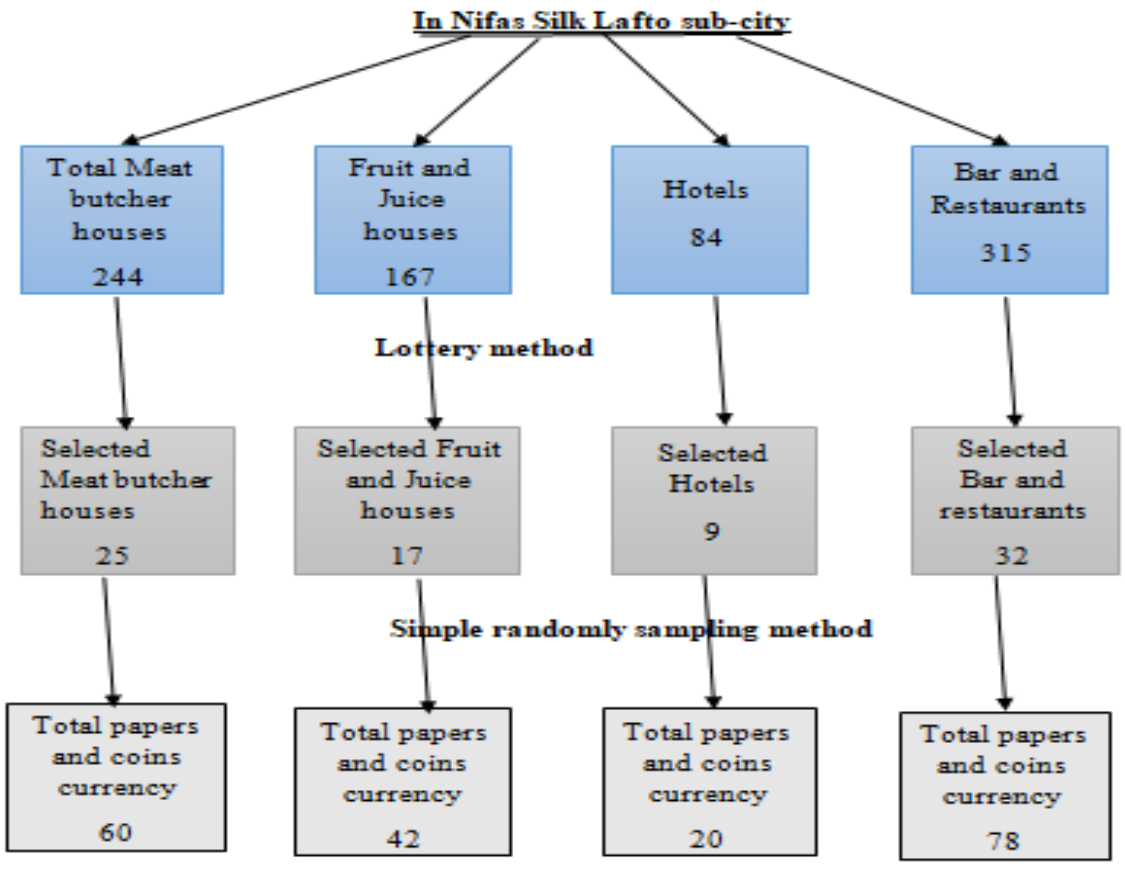

Figure 1 Schematic diagram of sampling method.

5.7. Measurement and Data collection

5.7.1. Data collection procedure

All samples were collected aseptically by letting the selected individuals to drop a selected currency note and coins into sterile polythene bags. The polythene bags were quickly sealed and the individuals were given a replacement equivalent to what they deposited in the polythene bags and data was collected. The polythene bags was immediately labeled and transported to Microbiology Laboratory, TASH for microbiological analysis. After that, a sterile cotton swab moistened with buffered peptone water solution is used for swabbing thoroughly on both surfaces of each sampled paper and coin currency placed on pre-sterilized aluminum foil this is larger than the size of paper notes and coin currencies and the swabs were one by one soaked into 10 ml sterile buffered peptone water solution. The samples were kept in refrigerator at 4°C until microbial analysis is conducted within one to two hours.

5.7.2. Laboratory analysis

Identification and Isolation of Bacteria

Isolation of various bacterial contaminants from the currency notes was performed via standard techniques described previously **[38, 39].** Briefly, after brought the soaked sample from refrigerator to room temperature and used a sterile cotton- tipped swab moistened with the previously soaked sample into 10 ml sterile buffered peptone water solution. The swabs were directly inoculated on MacConkey agar, Blood agar and chocolate agar. The pairs of inoculated media was incubated aerobically at 35-37^{0}C for 24 hours and then examined for bacterial growth. The isolated bacteria were then assessed for Gram reaction, and with the aid of using undertaking catalase and coagulase tests; hemolysis, sugar fermentation, and other biochemical tests, such as for citrate utilization, indole production, and urase activity; triple sugar iron (TSI) agar tests (for glucose, sucrose, and lactose fermentation); hydrogen sulfide (H_2S) and gas production tests; Motility and oxidase tests, according to CLSI protocols **[40, 41].**

Blood Agar Base preparation has been used as a base for making of blood agar and to help good growth of an extensive variety of fastidious microorganisms. Because it is an extremely nutritious medium it could additionally be used as a common purpose growth media without adding blood. The medium has sodium chloride for the osmotic balance. Blood Agar Bases are comparatively free of reducing sugars that have been reported to adversely impact the hemolytic reactions of beta-hemolytic streptococci. Sheep blood provides best outcomes for Group A Streptococci.

The low pH supports in stabilization of red blood corpuscles and favours the formation of clear haemolysiszone **[45].**

MacConkey Agar is a selective and differential medium. It is only slightly selective as the concentration of bile salts, which inhibit Gram-positive microorganisms, is low in contrast with other enteric plating media. Crystal violet also is included in the medium to inhibit the growth of Gram-positive bacteria, specially enterococci and staphylococci. Differentiation of enteric microorganisms is accomplished through the combination of lactose and the neutral red indicator. Colorless or pink to red colonies are produced relying upon the capability of the isolate to ferment the carbohydrate **[46].**

Antibiotic susceptibility was determined by the agar diffusion technique on Mueller- Hinton agar use including erythromycin 5µg, Cefoxitin 30µg, Gentamicin 10µg, penicillin 10IU, Clindamycin2µg, Tetracycline 30µg, Vancomycin 30µg, Ciprofloxacin 30µg, azithromycin 15µg, Ampicillin 10µg, Amikacin 30µg, Ceftazidime 30µg, Cephalothin 30µg, Trimethoprim+ sulfamethoxaz 1.25/23.75µg,Cefepime 30µg, meropenem (MEM) 10µg, and chloramphenicol (C) 30µg **[41- 44].** The Zone of growth inhibition was measured by using a caliper after 18–24hour incubation at 35–37°C. The diameters were interpreted according to the clinical and Laboratory Standards Institute (CLSI) guideline (29^{th} edition) as susceptible (S), intermediate (I), or resistant (R). The susceptibility and resistance was interpreted according to Clinical Laboratory Standard Institute (CLSI) guidelines (29^{th} edition). All laboratory activities were undertaken in the Microbiology laboratory **[41].**

5.8. Data Quality Assurance

A proactive process of managing quality is Quality Assurance. A quantity of measures were set during sampling, bacterial and statistical analysis to ensure making of quality results. Samples were put in sterile polythene bags and culturing was conducted under maximum aseptic conditions for example the media, wire loop, Petri dish all were sterilized before use. The working areas were disinfected with alcohol (70%) before and after working time. All petri-dishes were checked before use for sterility by incubating at 37°C then overnight and confirming for any microbial growths. To avoid errors Samples were labeled consequently increase accuracy. Laboratory standard operating procedures were followed to hold accuracy and consistency. Works were conducted under maximum supervision.

5.8.1. Data management and Quality control

Quality control is a reactive process with a set of activities used to confirm quality of the result. Data quality was ensured by using standardized data collection materials, proper training was given to all data collectors before the start of data collection, and intensive supervision during data collection by the principal investigator. During laboratory analysis pre-analytical, analytical and post-analytical stages of quality assurance which is incorporated in the Standard operating procedures (SOPs) of the microbiology laboratory of TASH was strictly followed. In addition, well-trained and experienced laboratory professionals were participate in the laboratory analysis procedure.

Pre-analytical phase

All steps of sample collection were strictly considered. All samples were collected aseptically by letting the selected individuals to drop separately a selected currency note and coins into sterile polythene bags to reduce cross contamination. The polythene bags were quickly sealed to reduce contamination. The polythene bags were immediately labeled and transported to Microbiology Laboratory of TASH for microbiological analysis. The swabs were one by one soaked into 10 ml sterile buffered peptone water solution. The samples were kept in refrigerator at 4°C until microbial analysis is conducted within one to two hours.

Analytical phase

At this stage of quality assurance all materials, equipment and procedures were adequately controlled. Culture media for its sterility, growth performance, stability and pH value was tested. To standardize the inoculum density of bacterial suspension for the susceptibility test, a McFarland standard equivalent to 0.5 was used. Standard reference strain S. aureus (ATCC-25923), E. coli (ATCC-25922) and P. aeruginosa (ATCC-27853) was also used as Control bacteria strains which was used in controlling the potency of the drugs. During all these steps a Standard operating procedures (SOPs) of the microbiology laboratory of TASH was strictly followed, and all the results were checked and approved by the supervisors.

Post-analytical phase

At this final stage of quality assurance, we were strictly followed the recording and documentation steps. The results were recorded with the sample code identification number. The terminology and format used in reporting was standardized. All reports were concise and clearly presented. Before leaving the microbiology laboratory, all reports were checked twice for correctness.

5.9. Data analysis and interpretation

Data was collected and entered into log book which was kept in a bookshelf. Computerized data was entered into excel software program application were keeping softcopy always. Data from questionnaires and microbial analysis was analyzed using statistical package for social science (SPSS) version 23. Comparison of data was carried out between various denominations of currency and statistical data analysis involved descriptive analysis of bacterial isolates, drug resistance pattern.

5.10. Operational definitions

Antimicrobial susceptibility: is stated as the highest dilution or lowest concentration of antibiotic that completely inhibits growth. **[47]**.

Bacterial contamination: is Presence of one or more potential food borne bacterial contaminants **[48]**.

Coin currency: is a piece of metal stamped and issued by the authority of a government for use as money **[49]**.

Currency: is a money system in use in a country, that in the form of paper or coins **[50]**.

Paper currency: is money issued by the government or the central bank as legal tender and which circulates as a substitute for specie **[51]**.

Clean currency: is the currency that seems clean when seen by naked eye (bare eye).

Dirty currency: is the currency that not clean and observed dirty thing from its surface with low degree when seen by naked eye (bare eye).

Very dirty currency: is the currency that not clean and observed dirty thing from its surface with high degree when seen by naked eye (bare eye).

Old currency: is the Ethiopian currency that currency used by current Ethiopian government without changing the currency that used during previous reign.

New currency: is Ethiopian currency that currency created and printed by current Ethiopian government by changing the currency that used during previous reign.

Food handlers: is the person who serves customers by providing foods at butchers, fruit and juice houses, bar and restaurants, and hotels while collecting currencies from customers for services they served.

5.11. Ethical considerations

The ethical approval and clearance was obtained from Addis Ababa University College of Health Science and Addis Ababa public Health Research and Emergency Management Directorate. Permission was also obtained from Addis Ababa City Administration food and drug administration, and Nifas Silk Lafto Sub City food and drug administration office. Informed consent was obtained from each study subjects prior to the administration of questionnaire and collecting currency after explaining the purpose of the study to respondent and also to hotels, food restaurants, and juice houses. The right to decline or to withdraw from the study at any stage without incurring any penalty was explained. Confidentiality and anonymous was kept by omitting their personal identifications such as names was not recorded. So that the instruments and procedure was not caused any harm to the study subject. The objective of the study was explained to respondents and written consent was obtained to provide accurate & honest response.

5.12. Dissemination of the result

The result of this study was presented and submitted to Addis Ababa University, College of Health Science, Department of Diagnostic and Public Health Microbiology, Addis Ababa City Administration Health Bureau, Addis Ababa public Health Research and Emergency Management Directorate, Addis Ababa FMHACA, Nifas Silk Lafto Sub City Health Office and Nifas Silk Lafto Sub City FMHACA. In addition, the study finding was disseminated and presented at local and international workshops and seminars. Manuscript was submitted to peer reviewed journals for possible publication.

6. Results

In this study 200 (two hundred) Ethiopian paper cash and coins were analyzed for bacterial contamination. Two hundred and eighty seven (287) different bacteria were isolated from one hundred and eighty three (183) currency notes and coins, giving percentage of contamination to be 91.5% (Fig.2).

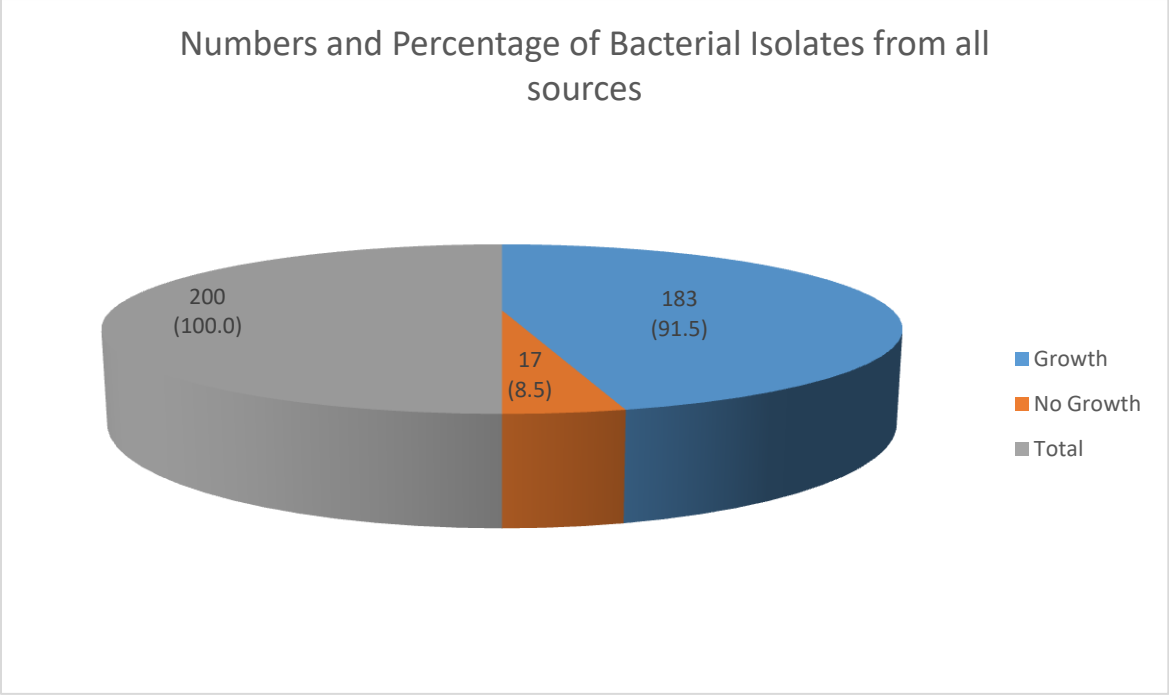

Figure 2: Numbers and percentage of bacterial isolates from all sources (n=200)

There were different currencies denominators collected from different collection sites. The highest number of currency collected were 10 ETB 27 (13.5%) from Fruit and Juice house, whereas the least number of currency collected were 1 ETB 1(0.5%) from Butcher (Table 1).

Table 1: Collection sites and distribution of currency notes

Collection site	Number of the currency notes (%)						
	1	5	10	50	100	200	Total
Bar and Restaurant	13(6.5)	12(6.0)	15(7.5)	16(8.0)	11(5.5)	5(2.5)	72(36.0)
Butcher	1(0.5)	3(1.5)	6(3.0)	3(1.5)	7(3.5)	5(2.5)	25(12.5)
Fruit and Juice house	16(8.0)	4(2.0)	27(13.5)	11(5.5)	4(2.0)	5(2.5)	67(33.5)
Hotel	6(3.0)	6(3.0)	6(3.0)	6(3.0)	6(3.0)	6(3.0)	36(18.0)
Total	36(18.0)	25(12.5)	54(27.0)	36(18.0)	28(14.0)	21(10.5)	200(100.0)

The different currency notes were classified based on their physical condition. There were most amount of dirty currency notes collected from fruit and juice houses (21.5%). The least amount of dirty currency notes and coins were collected from butchers (4.0%) (Table2).

Table 2: Physical condition of currency by sampling sources and currency type

		Quality status of the currency			
	Source/ currency	Clean No. (%)	Dirty No. (%)	Very Dirty No. (%)	Total
Sampling source	Bar and Restaurant	33(16.5)	39(19.5)	0(0.0)	72(36.0)
	Butcher	16(8.0)	8(4.0)	1(0.5)	25(12.5)
	Fruit and Juice house	23(11.5)	43(21.5)	1(0.5)	67(33.5)
	Hotel	13(6.5)	20(10.0)	3(1.5)	36(33.5)
	Total	85(42.5)	110(55.0)	5(2.5)	200(100.0)
Denomination	1 ETB	6(3.0)	29(14.5)	1(0.5)	36(18.0)
	5 ETB	2(1.0)	19(9.5)	4(2.0)	25(12.5)
	10 EBT	23(11.5)	31(15.5)	0(0.0)	54(27.0)
	50 ETB	18(9.0)	18(9.0)	0(0.0)	36(18.0)
	100 ETB	18(9.0)	10(5.0)	0(0.0	28(14.0)
	200 ETB	18(9.0)	3(1.5)	0(0.0)	21(10.5)
	Total	85(42.5)	110(55.0)	5(2.5)	200(100.0)

There were various bacterial growths from currency notes which appeared new and 'seemingly clean'. There were fourteen (14) different bacterial species isolated (Table 3). The most dominant bacteria isolated from the notes were coagulase-negative *staphylococci* 148 (51.60%) and the least isolated bacteria were *Acitinobacter* spp. 1 (0.30) (Table 3). However, no bacterial growth was observed in control currency. Coagulase-negative *staphylococci* was the most common bacterial isolates from 148 currency notes and coins of all denomination which were studied (Table 3).

Table 3: Numbers and percentage of bacterial isolates from all sources (n=200)

Type of isolated bacteria species	N (%)
Acitinobacter spp.	1(0.3)
Citrobactor spp.	10(3.5)
CoNS	148(51.6)
E.coli	4(1.4)
Entrobacter spp.	23(8.0)
Gram-positive Bacilli	35(12.2)
Klebsella spp.	11(2.8)
Proteus spp.	2(0.7)
Pseudomonas	4(1.4)
Salmonella spp.	4(1.4)
Serratia	7(2.4)
Shigella spp.	8(2.8)
Staphylococcus aureus	25(8.7)
Streptococcus spp.	5(1.7)
Total	287(100.0)

There were old and new Ethiopian currencies included in this study and were 56 (91.8%) old Ethiopian currencies contaminated with different bacteria, also there were 128 (92.1%) new Ethiopian currencies contaminated with different bacteria (Fig.3).

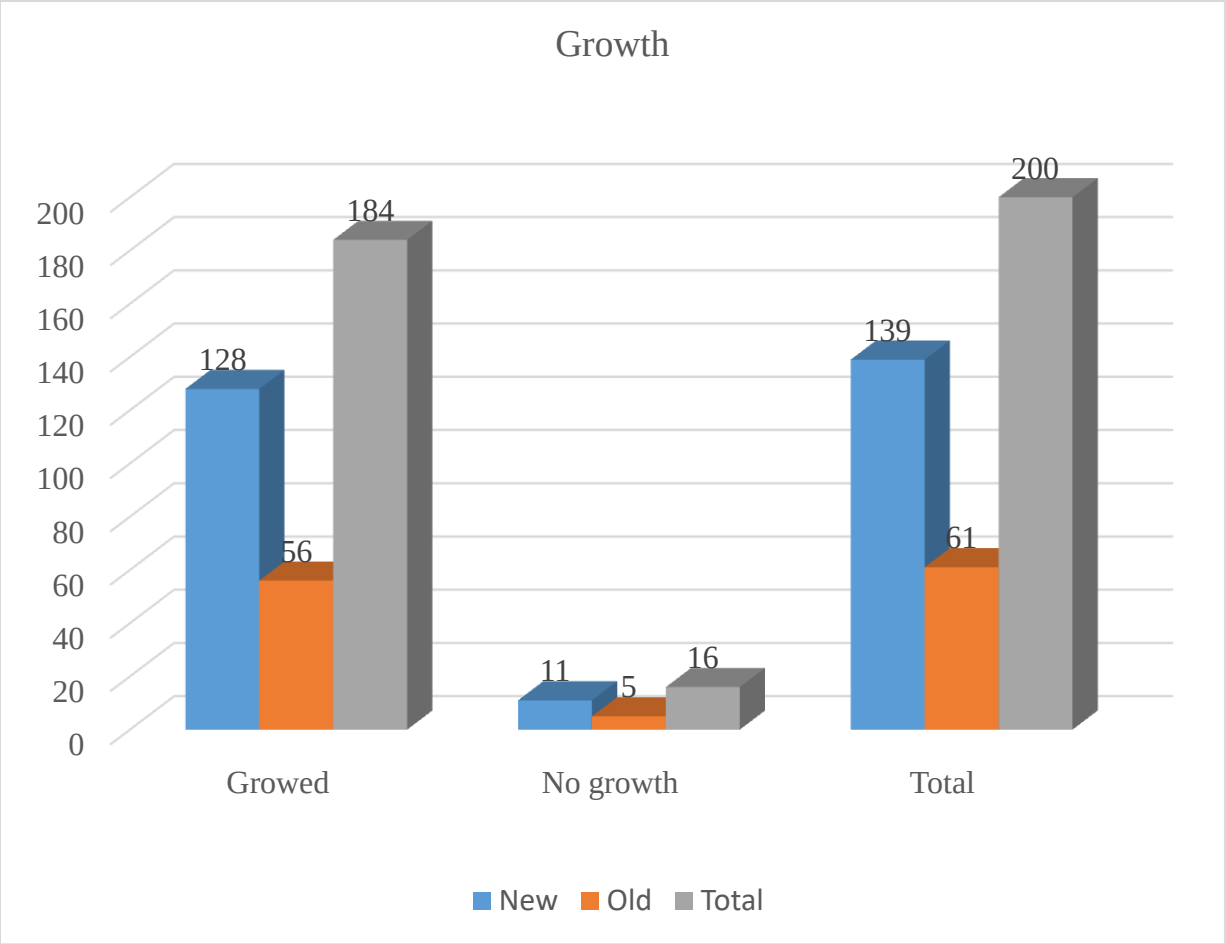

Figure 3: Numbers and percentage of Old and New currency contamination.

The highest bacterial contamination was 10 birr paper currency and the lowest bacterial contamination was detected in 200 birr paper currency (Table 4). Coagulase negative *staphylococci* there were relatively high bacterial contamination was 10 birr paper currency and 1birr coins (Table 4). There were relatively high Gram-positive *bacilli* contamination was with low denominator currency (10 birr, 5 birr and 1birr) (table 5). There were S. *auras* dominantly isolated from lower paper currency, However there were no isolation of S. *auras* from 1 birr coins (Table 4).

Table 4: Frequency Distribution of dominant bacteria among types of currency

Isolated Bacteria	N (%)						
	1Birr	5Birr	10Birr	50Birr	100Birr	200 Birr	Total
Acitinobacter spp.	0(0.0)	1(0.3)	0(0.0)	0(0.0)	0(0.0)	0(0.0)	0(0.3)
Citrobactor spp.	2(0.7)	1(0.3)	2(0.7)	2(0.7)	3(1.0)	0(0.0)	10(3.5)
CoNS	27(9.4)	16(5.6)	44(15.3)	25(8.7)	23(8.0)	13(4.5)	148(51.6.0)
E.coli	1(0.3)	0(0.0)	0(0.0)	3(1.0)	0(0.0)	0(0.0)	4(1.4)
Entrobacter spp.	0(0.0)	4(1.4)	9(3.1)	6(2.1)	4(1.4)	0(0.0)	23(8.0)
Gram-positive Bacilli	7(2.4)	7(2.4)	11(3.8)	4(1.4)	4(1.4)	2(0.7)	35(12.2)
Klebsella spp.	3(1.0)	0(0.0)	6(2.1)	1(0.3)	1(0.3)	0(0.0)	11(3.8)
Proteus spp.	0(0.0)	0(0.0)	2(0.7)	0(0.0)	0(0.0)	0(0.0)	2(0.7)
Pseudomonas	0(0.0)	0(0.0)	1(0.3)	2(0.7)	0(0.0)	1(0.3)	4(1.4)
Salmonella spp.	0(0.0)	0(0.0)	1(0.3)	1(0.3)	1(0.3)	1(0.3)	1(1.4)
Serratia	1(0.3)	2(0.7)	1(0.3)	0(0.0)	1(0.3)	2(0.7)	7(2.4)
Shigella spp.	2(0.7)	1(0.3)	2(0.7)	1(0.3)	1(0.3)	1(0.3)	8(2.8)
Staphylococcus aureus	0(0.0)	7(2.4)	6(2.1)	7(2.4)	1(0.3)	4(1.4)	25(8.7)
Streptococcus spp.	0(0.0)	0(0.0)	4(1.4)	0(0.0)	1(0.3)	0(0.0)	5(1.7)
Total	43(15.0)	39(13.6)	89(31.0)	52(18.1)	40(13.9)	24(8.4)	287(100.0)

There were relatively presence rates of Coagulase-negative *staphylococci* and Gram-positive *Bacilli s*pecies among currency notes isolated from fruit and juice house, and bar and restaurant whereas the isolation of Staphylococcus *auras* in currency notes collected from butcher and fruit and juice house were more dominant (Table 5).

Table 5: Frequency Distribution of dominant bacteria among collection sites

Isolated Bacteria	N (%)				
	Bar and Restaurant	Butcher	Fruit and Juice house	Hotel	Total
Acitinobacter spp.	0(0.0)	1(0.3)	0(0.0)	0(0.0)	1(0.3)
Citrobactor spp.	2(0.7)	0(0.0)	4(1.4)	4(1.4)	10(3.5)
CoNS	58(20.2)	15(5.2)	49(17.1)	26(9.1)	148(51.6)
E.coli	1(0.3)	0(0.0)	3(1.0)	0(0.0)	4(1.4)
Entrobacter spp.	5(1.7)	3(1.0)	14(4.9)	1(0.3)	23(8.0)
Gram-positive Bacilli	14(4.9)	1(0.3)	13(4.5)	7(2.4)	35(12.2)
Klebsella spp.	1(0.3)	2(0.7)	8(2.8)	0(0.0)	11(3.8)
Proteus spp.	1(0.3)	0(0.0)	1(0.3)	0(0.0)	2(0.7)
Pseudomonas	2(0.7)	2(0.7)	0(0.0)	0(0.0)	4(1.4)
Salmonella spp.	0(0.0)	4(1.4)	0(0.0)	0(0.0)	4(1.4)
Serratia	2(0.7)	0(0.0)	3(1.0)	2(0.7)	7(2.4)
Shigella spp.	4(1.4)	1(0.3)	2(0.7)	1(0.3)	8(2.8)
Staphylococcus aureus	3(1.0)	8(2.8)	11(3.8)	3(1.0)	25(8.7)
Streptococcus spp.	0(0.0)	1(0.3)	4(1.4)	0(0.0)	5(1.7)
Total	93(32.4)	38(13.2)	112(39.0)	44(15.3)	287(100.0)

There were different gram positive and gram negative bacteria resistant with different antibiotics (Table 6-8). The highest resistance rates in gram-positive bacteria were against penicillin, erythromycin, ampicillin and clindamycin (Table 6) whereas ampicillin was the most resistance gram-negative bacteria (Table 7).

Table 6: Antimicrobial Susceptibility Patterns of gram-positive Bacteria Isolates

Antimicrobial Agent	Staphylococcus aureus			Coagulase Negative Staphylococcus			Streptococcus spp.		Gram-positive Bacilli (Bacillus)		
	N (%)S	N (%)R	N (%)I	N (%)S	N (%)R	N (%)I	N (%)S	N (%)R	N (%)S	N (%)R	N (%)I
FOX (Cefoxitin)	22 (88.0)	3 (12.0)	0	97 (65.5)	51 (34.5)	0	-	-	23 (65.7)	12 (34.3)	0
Gentamycin	21 (84.0)	3 (12.0)	1 (4.0)	135 (91.2)	10 (6.8)	3 (2.0)	5 (100.0)	0	32 (91.4)	2 (5.7)	1 (2.9)
Penicillin	19 (76.0)	6 (24.0)	0	75 (50.7)	73 (49.3)	0	4 (80.0)	1 (20.0)	17 (48.6)	18 (51.4)	0
Clindamycin	13 (52.0)	12 (48.0)	0	73 (49.3)	65 (43.9)	10 (6.8)	4 (80.0)	1 (20.0)	18 (51.4)	13 (37.1)	4 (11.4)
Erythromycin	19 (76.0)	4 (16.0)	2 (8.0)	87 (58.8)	45 (30.4)	16 (10.8)	2 (40.0)	3 (60.0)	16 (45.7)	16 (45.7)	3 (8.6)
Tetracycline	21 (84.0)	4 (16.0)	0	102 (68.9)	35 (23.6)	11 (7.4)	5 (100.0)	0	28 (80.0)	3 (8.6)	4 (11.4)
Amoxicillin	19 (76.0)	6 (24.0)	0	111 (75.0)	21 (14.2)	16 (10.8)	3 (600.0)	2 (40.0)	14 (40.0)	15 (42.9)	6 (17.1)
Ampicilin	19 (76.0)	6 (24.0)	0	74 (50.0)	74 (50.0)	0	4 (80.0)	1 (20.0)	17 (48.6)	19 (51.4)	0
Ciprofloxacin	21 (84.0)	4 (16.0)	0	111 (75.0)	26 (17.6.0)	11 (7.4)	5 (100.0)	0	28 (80.0)	5 (14.3)	2 (5.7)

N: Number of Sensitive, Number of Resistant and Number of Intermediate: %S: Percentage Sensitive: %R: percentage Resistant: %S: Percentage Intermediate.

Table 7: Antimicrobial Susceptibility Patterns of gram-negative Bacteria Isolates

Antimicrobial Agent	Acitinobacter spp.		Citrobactor spp.			E.coli		Entrobacter spp.			Klebsella spp.	
	N (%)S	N (%)R	N (%)S	N (%)R	N (%)I	N (%)S	N (%)R	N (%)S	N (%)R	N (%)I	N (%)S	N (%)R
Gentamycin	1 (100.0)	0	10 (100.0)	0	0	3 (75.0)	1 (25.0)	23 (100.0)	0	0	0	11 (100.0)
Ampicillin	0	1 (100.0)	4 (40.0)	6 (60.0)	0	0	4 (100.0)	4 (17.4)	19 (82.6)	0	0	11 (100.0)
Amoxicillin	0	1 (100.0)	1 (10.0)	9 (90.0)	0	2 (50.0)	2 (50.0)	9 (39.1)	14 (60.9)	0	5 (45.5)	6 (54.5)
Chloramphenicol	1 (100.0)	0	10 (100.0)	0	0	3 (75.0)	1 (25.0)	22 (95.7)	1 (4.3)	0	10 (90.9)	1 (9.1)
Cephalothin	1 (100.0)	0	6 (60.0)	4 (40.0)	0	3 (75.0)	1 (25.0)	19 (82.6)	4 (17.4)	0	9 (81.8)	2 (18.2)
Cefazolin	1 (100.0)	0	9 (90.0)	1 (10.0)	0	3 (75.0)	1 (25.0)	20 (87.0)	3 (13.0)	0	11 (100.0)	0
Amikacin	1 (100.0)	0	10 (100.0)	0	0	4 (100.0)	0	21 (91.3)	1 (4.3)	1 (4.3)	10 (90.9)	1 (9.1)
Trimethoprim +sulfamethoxaz	1 (100.0)	0	10 (100.0)	0	0	4 (100.0)	0	21 (91.3)	2 (8.7)	0	11 (100.0)	0
Cefepime	1 (100.0)	0	5 (50.0)	3 (30.0)	2 (20.0)	4 (100.0)	0	21 (91.3)	2 (8.7)	0	10 (91.0)	1 (9.0)
Ceftazidine	1 (100.0)	0	5 (50.0)	5 (50.0)	0	2 (50.0)	2 (50.0)	17 (73.9)	6 (26.1)	0	9 (81.8)	2 (18.2)
Meropenem	1 (100.0)	0	10 (100.0)	0	0	3 (75.0)	1 (25.0)	22 (95.7)	1 (4.3)	0	11 (100.0)	0

N: Number of Sensitive, Number of Resistant and Number of Intermediate: %S: Percentage Sensitive: %R: percentage Resistant: %S: Percentage Intermediate.

Table 8: Antimicrobial Susceptibility Patterns of gram-negative Bacteria Isolates

Antimicrobial Agent	Proteus spp.		Pseudomonas		Salmonella spp.		Serratia			Shigella spp.		
	N (%)S	N (%)R	N (%)S	N (%)R	N (%)S	N (%)R	N (%)S	N (%)R	N (%)I	N (%)S	N (%)R	N (%)I
Gentamycin	2 (100.0)	0	4 (100.0)	0	-	-	7 (100.0)	0	0	8 (100.0)	0	0
Amoxicillin	1 (50.0)	1 (50.0)	-	-	1 (25.0)	3 (75.0)	2 (28.6)	5 (71.4)	0	3 (37.5)	5 (62.5)	0
Ampicillin	2 (100.0)	0	0	4 (100.0)	3 (75.0)	1 (25.0)	3 (42.9)	4 (57.1)	0	2 (25.0)	6 (75.0)	0
Chloramphenicol	2 (100.0)	0	3 (75.0)	1 (25.0)	4 (100.0)	0	6 (85.7)	1 (14.3)	0	5 (62.5)	1 (12.5)	2 (25.0)
Cephalothin	1 (50.0)	1 (50.0)	3 (75.0)	1 (25.0)	1 (25.0)	3 (75.0)	7 (100.0)	0	0	3 (47.5)	5 (62.5)	0
Cefazolin	1 (50.0)	1 (50.0)	-	-	2 (50.0)	2 (50.0)	5 (71.4)	2 (28.6)	0	4 (50.0)	4 (50.0)	0
Amikacin	2 (100.0)	0	4 (100.0)	0	4 (100.0)	0	7 (100.0)	0	0	8 (100.0)	0	0
Trimethoprim sulfamethoxaz	2 (100.0)	0	4 (100.0)	0	4 (100.0)	0	5 (71.4)	1 (14.3)	1 (14.3)	8 (100.0)	0	0
Cefepime	2 (100.0)	0	1 (25.0)	3 (75.0)	4 (100.0)	0	3 (42.9)	2 (28.6)	2 (28.6)	3 (37.5)	4 (50.0)	1 (12.5)
Ceftazidine	2 (100.0)	0	1 (25.0)	3 (75.0)	4 (100.0)	0	3 (42.9)	4 (47.1)	0	4 (50.0)	4 (50.0)	0
Meropenem	2 (100.0)	0	3 (75.0)	1 (25.0)	4 (100.0)	0	5 (71.4)	2 (28.6)	0	5 (62.5)	2 (25.0)	1 (12.5)

N: Number of Sensitive, Number of Resistant and Number of Intermediate: %S: Percentage Sensitive: %R: percentage Resistant: %S: Percentage Intermediate.

7. Discussion

Money are commonly handled by numerous people with different hygienic conditions, are also placed in different environments, and therefore Cash-money considered as an important reservoir of various groups of pathogenic bacteria for the transmission within community. The results of our study indicated that 91.5% of our studied Ethiopian money was contaminated with different types of bacteria. Similarly, various kinds of bacteria were reported from money of Saudi Arabia 88% **[3]**, Ethiopia 98.5% **[5]**, South Africa 96% **[7]**, India 90% **[13]** Iran 77.7% **[52]**, Bangladesh 94% **[53]**, Ghana 97.1% **[54]**. In this study bacterial pathogens such as gram positive bacilli, S. aureus, Entrobacter, Citrobactor, Klebsella, Shigella, Serratia, Streptococcus, E. coli, Salmonella, Pseudomonas, Proteus, and Acitinobacter were isolated ETB samples of "1 ETB coin", 5 ETB note", "10 ETB note", "50 ETB", "100 ETB" and "200 ETB". Similarly, various types of bacterial pathogenic were reported from money of Saudi Arabia **[3]**, Ghana **[4]**, Ethiopia **[5, 6]**, South Africa **[7]**, India **[8]**, and Bangladesh **[12]**, and Rwanda **[15]**, However the load and survival rate of bacteria on the cash-money of different countries varies **[55]**. Ethiopian currency collected from different sites at South western, south eastern, Easter, and western part of the country were highly contaminated with bacteria **[5, 6, 9, 54]**.

According to this study indicated Ethiopian cash-money were contaminated with Staphylococcus *aureus* 8.7% similarly were reported from currency of Saudi Arabia 13% **[3],** Ghana 7.14% **[4],** South Africa 11% **[7],** Bangladesh 4.2% **[12],** India 4.2% **[21]**, and Tanzania 7.40% **[33].** whereas lower than the percentage of cash- money contamination study reported from Bangladesh 25% **[12]** and Zambia 25.85% **[36].** Also this study shows 12.2% ETB were contaminated by Gram-positive *Bacilli*, similar study conducted in Bangladesh shown 9% **[12],** Sudan 12.7% **[24]** and Nigeria 10.1% **[28].** There were 2.8% ETB currency contaminated by *Klebsella spp*. were similar to study reported in Colombia 1.6% **[31],** Tanzania 3.84% **[33],** Ghana 3.51%**[4],** Tanzania 7.40% **[33]** and Ghana 5.6% **[30].** In this study currency contamination by *Pseudomonas* spp. were 1.4%. Whereas another study shown in Jimma, Ethiopia 0.62% **[5],** Bangladesh 0.8% **[12],** India 0.8% **[21]** and Tanzania 7.40% **[33].** According to this study *Acinetobacter spp.* 0.3%. While study indicated in Jimma 1.45% **[5]** and Colombia 3.2% **[31].** Currency contaminated with *E. coli* were 1.4%. Similarly reported from Saudi Arabia 2% **[3],** Ghana 1.75% **[4],** Nigeria 3.8% **[28]** and Colombia 3.2% **[31].** In our study the *Coagulase-negative staphylococci* contamination were 51.6%. The other study shown in Saudi Arabia 64% **[3],** and Ghana 38.59% **[4].** Also this study indicated 3.5% *Citrobacter spp.*

contamination. Whereas study reported in Ghana 3.51% **[4],** South Africa 5.8% **[7],** Sudan 2% **[24].** According to this study cash-money contamination by *salmonella spp.* 1.4%. Similar study shown in South Africa 2.5%) **[7]** and Sudanese 3.9% **[24].** The contamination of *Shigella spp.* in this study was 2.8%. Similarly study conducted in Ghana 2.68% **[4],** South Africa 1.7% **[7]** and India 0.8% **[21].** According to this study indicted currency contaminated by *Entrobacter spp.* 8.0%. Also as shown study done in Bangladesh 19.2% **[12],** India 19.2% **[21]**, Nigeria 8.9% **[28],** Ghana 2.8% [**30]** 2.8% and Colombia 2.8% **[31].** In this study there were 0.7% of currency contaminated by proteus spp. whereas reported from study conducted Bangladesh l.7% **[12]**, India l.7% **[21]**, and Ghana 8.4% **[30]**.

The different money used in this study indicated different degree of contamination with some denominations having high counts and others lower counts. However, there were high variations that bacterial load within samples of the same denominator money or the same collection site. Most of the money were dirties and it most contaminated with various bacteria load, particularly lower denominations money of 1 ETB (coin), 5 ETB (note), and 10 ETB (note). However there were 1 ETB (coins) the lowest bacterial contamination when compared with 5 ETB (note), and 10 ETB (note). A small number of studies have examined the contamination of coin currency, and copper seems to be a limiting factor for bacterial survival on coins **[56].** Accordingly, coins were found to carry opportunistic bacterial pathogens, but they exhibit a lower bacterial load than paper currency **[56]**.

As indicated in this study Coins have been shown to carry potential pathogens and opportunistic pathogens bacteria, such as a variety of species of the *CoNS,* Gram-positive *Bacilli, Citrobactor spp., E.coli, Klebsella spp., Serratia, and Shigella spp*. were isolated. In this study there were no growths of streptococcus spp. from coins. The absence of streptococci isolates from coins probably suggests a high sensitivity of these bacteria to coins (metallic Cu) [56].

Generally the bacterial counts were related to the physical condition of the money as shown in the high bacterial load in 1 ETB, 5 ETB, and 10 ETB. These are among the currency notes which are frequently used in daily money transactions in markets with small business. In compare, the cash-money which is not commonly used for daily activities like, 200 ETB had least bacterial contamination. This also depends on how long money stayed in circulation. The source of contamination of the circulating currencies would be due to contamination during circulation and handling as no bacteria were growth from new cash-money used as controls.

In this study all paper money of Ethiopia was included. However, only 1ETB coin selected to the study and the other coin cents not included due to depletion from the market. Coins were not sampled because their circulation rate is too low due to the depletion of currency value in Nepal **[11]**.

This study evidently indicates the Ethiopian paper money and coin currencies are contaminated with pathogenic bacteria and it shows the potential for paper notes and coins to spread pathogenic bacteria in the Ethiopian community. The study checks how essential it is for food handlers to abstain from handling cash, since the oral cavity is a main portal for transition of bacterial pathogens. It is of dominantly importance the general population, and especially food handlers, wash their hands thoroughly after handling cash-money and before handling food. Those Customers with unclean hands could additional increase their risk of contracting food related diseases **[57].**

This study confirmed the existence of antibiotic resistant bacteria and there were different types of bacteria were resistant to different antibiotics. Additionally, many of these isolates were resistant to multiple antibiotics, which is a threat to public health in Ghana **[58]**. There is an increasing threat of antibiotic resistance due to excessive usage without prescriptions from a pharmacist **[59, 60]**.

Antibiotics Susceptibility Pattern was resistant to various Gram-negative bacteria species. Thus, Ampicillin and Gentamicin 100% resistant to Klebsella spp. similarly Ampicillin 100% resistant to Pseudomonas. Whereas Amoxicillin 90.0% resistance to Citrobactor spp. and Ampicillin 82.6%, 75.0% resistant to Entrobacter spp. and Shigella spp. respectively. However, Meropenem, Cefazolin and Trimethoprim+ sulfamethoxaz were 100% sensitive klebsella spp. similarly Gentamycin, Chloramphenicol, Amikacin, Trimethoprim+sulfamethoxaz and Meropenem were sensitive to Citrobactor spp. also E.coli 100% sensitive to Amikacin and Trimethoprim + sulfamethoxaz, whereas Gentamycin 100% sensitive to Entrobacter spp. additionally, Gentamycin, Amikacin and Trimethoprim + sulfamethoxaz were 100% Shigella spp., also there were Meropenem, Cefepime, and Ceftazidine 100% sensitive to *Salmonella spp*.

Additionally Antibiotic Susceptibility pattern of the isolated bacteria against selected antibiotics, there were for gram positive bacteria isolates showed 55.12%, 46.01%, 42.72%, 31.92%,

31.73%, 31.80%, 19.72%, 16.43% and 9.54% resistant to ampicillin, penicillin, clindamycin, erythromycin, cefoxitin, amoxicillin, tetracycline, Ciprofloxacin and gentamycin respectively. Also 8.11%, 28.38%, 20.00%, 2.70%, 4.05%, 20.27%, 35.14% and 9.46 were resistant to Chloramphenicol, Cephalothin, Cefazolin, Amikacin, Trimethoprim+sulfamethoxaz, Cefepime, Ceftazidine and Meropenem respectively. Also in this study Methicillin-resistant Staphylococcus aureus (MRSA) was found to be 12% using cefoxitin. Whereas study conducted in South Africa shown MRSA was 36.4%.

Similarly, Study from Ghana showed varied resistance to commonly use antibiotics with Coagulase negative *staphylococci*, *Streptococcus spp*.,where also 87.5% Isolates resistance to *Salmonella* and *Entrobecters* spp., were also 100% resistant to Ampicillin and Penicillin **[29].** Whereas S. aureus was 75.0% resistant **[29].** Additionally as study indicated in Nigeria isolated bacteria were 100% resistant to Amoxicillin, 87.5% resistant to tetracycline and chloramphenicol, and 50% resistant to Gentamycin **[29].**

As study conducted in South Africa, S. aureus resistant range of between 62.5 and 100%. Similarly study reported from Indian currency, 100% penicillin ressistance to S. aureus. Whereas 100% gentamicin sensitive to Acinetobacter spp., citrobactor spp, klebsiella pps, and Pseudomonas **[34]**.

Paper currency notes collected from Tanzania shows that 28.125% S.aureus isolates were multidrug resistant **[35].** This study supported by study conducted in Zambia, Paper currency notes were collected from restaurants and hotels and the prevalence of S.aureus is 25.85% **[36].** Drugs would have been seriously abused by the people due to self-medication and over-dose **[61, 62]**. This difference most probably because of misuse drug, types of currency and geographical difference.

As our study indicated that the Ethiopian cash-money was seriously contaminated with potential bacterial isolates that were highly resistant to the most broadly used available antibiotics. Generally this study has demonstrated that the Ethiopian currencies notes were heavily contaminated with highly potential bacterial isolates that are extremely resistant to the most broadly used antibiotics and are a threat to public health. Therefore, there is need to give attention misuse drugs. Ethiopian cash-money was critically abused with dirty and unhygienic hands contaminating them with pathogenic bacteria with the potential of infecting handlers. Therefore, there is need to educate the community on the effect of improper handling of ETB and women most especially because of their children who put money in their mouth.

8. Strength and Limitation

8.1. Strength

- All woreda in NSL sub city were included in the study.

8.2. Limitation

- The risk factors of currency contamination were not included in the study.
- Different denomination of currency not equally collected from different source due to absence of denomination from food handlers.

9. Conclusion and Recommendation

9.1. Conclusion

In this study the isolation of bacteria from Ethiopian paper money and coins currency reported that confirmed currency might be a vector playing an important role in the transmission of pathogenic bacteria in the community. The isolated bacteria such as Coagulase negative staphylococci, gram positive bacilli, S. aureus, Entrobacter, Citrobactor, Klebsella, Shigella, Serratia, Streptococcus, E. coli, Salmonella, Pseudomonas, Proteus, and Acitinobacter that cause disease in healthy individuals and opportunistic pathogens that may cause disease in hospitalized and immunocompromized patients. Potentially pathogenic bacteria that are highly resistant to the most widely used antibiotics and are a threat to public health. Therefore handling of paper currency and coins deserve special attention. The practice of licking or applying saliva to the fingers while counting money is an important potential route of exposure to enteric pathogen. The improper handling (or, abuse of) currency notes should, therefore, be seen as a dangerous habit or practice. The evidence for the presence of pathogenic bacteria on money reinforces the need for strict hygienic practices among money handlers who also handle food and water.

9.2. Recommendations

- The government could create awareness and give health education to community about transmission of potential infection during cash circulation.
- Food handlers, wash their hands thoroughly after handling cash-money and before handling food.
- Ethiopian National bank Promote father strengthening the usage of online shopping, Credit Cards and using money transaction technology (like mobile banking) instead of cash contact by hands.
- Regulatory body could work on drug administration for manage drug resistance

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11. Annex

11.1. Annex I Structured English Version Questionnaire Information form

Code No._____

Hello Ms. My name is ---------- am here on behalf of Hiko Negeho student of Addis Ababa University College of Health Sciences Department of Medical Laboratory Sciences. We are conducting a study on Magnitude of Bacterial contamination of Ethiopian paper notes and coin currency and their antimicrobial susceptibility pattern from food handlers in Nifas Silk Lafto Sub-City. You are invited to take part in a research study.

Before you decide whether to participate, you need to understand, why the research is being done and what it would involve. Please take the time to listen as I read the following information.

Please ask me if there is anything that is not clear, or if you would like more information. When all of your questions have been answered and you feel that you understand this study, you will be asked if you wish to participate in the study.

Purpose of the Study and Study Requirements the study entitled "Magnitude of Bacterial contamination of Ethiopian paper notes and coin currency and their antimicrobial susceptibility pattern from food handlers in Nifas Silk Lafto Sub-City, Addis Ababa, Ethiopia".

According to the research eligibility, criteria you are selected as one of study participant by chance as you are food handler from selected meat butchers houses/juice houses/hotels/food restaurants. The study will help us to know Magnitude of Bacterial contamination of Ethiopian paper notes and coin currency and their antimicrobial susceptibility pattern from food handlers.

If you agree to take part in the study, you will also be asked to answer questions in relation to your exchange paper note and coin currencies. This will take you about 4 minutes.

Risks- An inconvenience may be the time and effort you take to be a participant. You may find one or more questions that we ask to be upsetting or emotionally sensitive. You do not have to respond to any question that makes you uncomfortable.

Benefits- There is no direct benefits to you for participating in the study. You may find an indirect benefit in knowing you have participated in an important study that could help others in the future

Voluntariness- Your participation in this study is voluntary. If you decide not to participate, you will not lose any existing benefits to which you are entitled. If you agree to participate in this

study, you may end your participation at any time without penalty or loss of existing benefits to which you are entitled. If you decide to take part, you are free to skip any questions. You are free to withdraw at any time without affecting your relationship with the service providers.

11.2. አባሪ ፤ የተሳታፊውን ፈቃድ መግለጫ (Consent form Amharic version)

ኮድ ቁጥር _____

ሰላም፤ እኔ ስሜ---------- እኔ እዚህ የተገኘሁበት ለሂኮ ነገሆ የአዲስ አበባ ዩኒቨርሲቲ የጤና ሳይንስ ኮሌጅ የህክምና ሳይንስ ላቦራቶሪ ክፍል ተማሪ ነው።

እኛ በንፋስ ስልክ ላፍቶ ክፍለ ከተማ የኢትዮጵያን ቢር (ገንዘብ) ላይ የሚገኝ በሽታ አምጪ ተዋሲያን (ባክቴሪያ) መጠን እና የመድኃኒት መላመድን ከምግብ አቅራቢዎች በመሰበሰብ ጥናት ዕያካሄደን ነው።

ጥናቱ ውስጥ ላይ ከመሳተፍህ/ሽ በፊት የጥናቱ አስፈላጊነትና የሚያሳትፈው መረዳት አለብህ/ሽ. የሚቀጥለሁን ሃሳብ አነብለሀለሁ እባክህ/ሽ ትንሽ ጊዜ ወሰደህ/ሽ አደምጠኝ/ጪኝ።

እባክህ/ሽ ግለጽያሎነልህ/ሽ ነግር ካለ እና ተጨማሪ ማብራሪያ ከፈለክ/ሽ ጠይቀኝ/ቂኝ። ሁሉም ጥያቄዎችህ/ሽ ከተመለሱልህ/ሽ እና ስለጥናቱ ከተረዳህ/ሽ በኋላ ጥናቱ ውስጥ መሳተፍ ፍላጎት ካለህ/ሽ ትጠየቃለህ/ሽ።

የጥናቱ አላማና ለጥናቱ የሚያስፈልግ በኢትዮጵያ፣ አዲስ አበባ፣ ንፋስ ስልክ ላፍቶ ክፍለ ከተማ የኢትዮጵያን ቢር (ገንዘብ) ላይ የሚገኝ በሽታ አምጪ ተዋሲያን (ባክቴሪያ) መጠን እና የመድኃኒት መላመድን ከምግብ አቅራቢዎች በመሰበሰብ ማጥናት ላይ የተፈቀደው።

በጥናት መስፈርት መሙያ መሰረት የጥናቱ ተሳታፊ እንድትሆን/ኚ እንደ ምግብ አቅራቢነትህ/ሽ ከተመረጡ ስጋ ቤቶች፣ ጁስ ቤቶች፣ ሆቴሎች እና የምግብ ሬስቶራንቶች በእጣ ተመርጠሀል/ሻል። ጥናቱ የኢትዮጵያን ቢር (ገንዘብ) ላይ የሚገኝ በሽታ አምጪ ተዋሲያን (ባክቴሪያ) መጠን እና የመድኃኒት መላመድን ከምግብ አቅራቢዎች በመሰብሰብ እንድናቀው ይረዳናል።

ጥናቱ ውስጥ ለመሳተፍ ፍቃደኛ ከሆንክ/ሽ ስለ ብር መቀያየር ጋር የተያያዘው መልስ እንድትሰጥ/ጪ ትጠየቃለህ/ሽ። ይህ ወደ 4 ደቂቃ ይፈጃል።

ስጋቶች- ጥናቱ ላይ በምትሳተፍበት/ፊበት ጊዜ የማይመችህ/ሽ ጥያቄ (ሀሳብ) ከጠየቀንህ/ሽ ምላሽ ላትስጥ/ጪ።

ጥቅሞች- ይህንን ጥናት ውስጥ ስለተሳተፍክ/ሽ ቀጥታ ጠቀሜታ የለውም። ግን በተዘዋዋሪ በዚህ ጠቃሚ ጥናት ውስጥ በመሳተፍክ/ሽ ወደፊት ለሌሎች ይጠቅማል።

ፍቃደኝነት- በጥናቱ ተሳትፎህ/ሽ በፍቃደኝነትህ/ሽ ላይ የተመሰረታ ንው። ለመሳተፍ ፍቃደኛ ካልሆክ/ሽ የትኛውም የለውን ጥቅም ማጣት የለብህም/ሽም ባንተህ/ቺ ፍቃደኝነት ይሆናል። በጥናቱ ለመሳተፍ

ፍቃደኛ ከሆክ/ሽ በየትኛውም ጊዜ ያለምንም ቅጣት ተሳትፎህን/ሽን ማቋረጥ ትችላለህ/ሽ. ከተሳተፍክ/ሽ የትኛውም ጥያቄ አለመመለስ መብትህ/ሽ ነው ምንም አይነት የግኙኝነት ችግር አይፈጥም።

Name of principal investigator: **Hiko Negeho**

Address- Department of Medical Laboratory Sciences, College of Health Science, Addis Ababa University, Addis Ababa, Ethiopia. Cell phone: 913412346 e-mails:hikonegeho@gmail.com

የዋና ተመራማሪው ስም፡ ሂኮ ነገሆ

አድራሻ፤ የሕክምና ላቦራቶሪ ቴክኖሎጂ ዲፓርትመንት፣ የጤና ሳይንስ ኮሌጅ፣ አዲስ አበባ ዩኒቨርሲቲ- አዲስ አበባ፣ ኢትዮጵያ። ስልክ ቁጥር፡ 251913412346

ኢ-ሜይል፡hikonegeho@gmail.com

Informed Consent

You have been already briefly informed about the study and clearly understand the objective.

Now please tell me would you be willing to participate in the study? 1. Agree 2. Disagree

1. Agreed, Thanks! Conduct the interview
2. Did not agree, Thanks! Proceed to the next eligible participant

Print name of participant, date and signature or thumb impression of participant

_____ ____ /____ /____ (dd/mm/yy)

Date of interview (Ethiopian calendar) _____/_____/____

Print name of researcher, date and signature of researcher

_____ _____/____/_____ (dd/mm/yy)

የፍቃደኝነት መሳወቅያ

በአጭሩ ስለ ጥናቱ አላማ ተገልጾለሃል/ሻል በደንብ ተግንዝበሀል/ሻል። እባክህ/ሽ አሁን ንገረኝ/ሪኝ ጥናቱ ላይ ለመሳተፍ ፍቃደኛነህ/ሽ? 1. አዎ 2. አይደለሁም

1. ተስማምቻለሁ፣ አመሰግናለሁ! ቃለመጠይቅ ይደረግ።
2. አልተስማመሁም፣ አመሰግናለሁ! ወደሌለው የጥናት ተሳታፊ እለፍ።

የተመራማሪ ስም፣ ቀን ና ፍርማ

_____ ___ /___ /___ (ቀን/ወር/ዓ.ም)

የቃለ መጠየቅ ቀን (በኢትዮጵያ አቆጣጠር)

የዋና ተመራማሪው ስም፣ ቀን ና ፍርማ

_____ ____ /____ /____ (ቀን/ወር/ዓ.ም)

11.3. Annex II Questioner on Magnitude of bacterial contamination of Ethiopian paper money and coin currency and their antimicrobial susceptibility test from food handlers in NSL SC, AA, Ethiopia, May, 2021.

Q.no	Questionnaires	Response	Skip
101	Sample source	A. Meat butcher B. Fruit and juice house C. Hotel D. Food restaurant	January 15, 2021 to May 30, 2021
102	Denomination of currency	A. 1 Birr B. 5 Birr C. 10 Birr D. 50 birr E. 100 Birr F. 200 birr	
103	Currency condition	A. Old B. New	
104	Physical status of currency	A. Clean B. Dirty C. Very dirty	
105	Types of Antibiotics	A. erythromycin 5µg B. Cefoxitin 30µg C. Gentamicin 10µg D. penicillin 10IU E. Clindamycin 2µg F. Tetracycline 30µg G. Ciprofloxacin 30µg H. Amoxicillin 30µg I. Ampicillin-G 10µg J. Amikacin 30µg K. Ceftazidime 30µg	

		L. Cephalothin 30µg M. Cefazolin N. Trimethoprim+ sulfamethoxaz 2.25/23.75µg O. Cefepime 30µg P. Meropenem 10µg Q. Chloramphenicol 30µg	

11.4. አባሪ II የኢትዮጵያን ብር (ገንዘብ) ላይ የሚገኝ በሽታ አምጪ ተዋሲያን (ባክቴሪያ) መጠን አና የመድኃኒት መላመድን ከምግብ አቅራቢዎች በመሰበሰብ ማጥናት በንፋስ ስልክ ላፍቶ ክፍለ ከተማ፣ አዲስ አበባ፣ ኢትዮጵያ፣ ግንቦት፣ 2021.

ተ.ቁ	ጥያቄዎች	ምላሽ	ይለፈኝ
101	የናሙና ምንጭ	ሀ. ስጋ ቤት ለ. ፍራፍሬና ጁስ ቤት ሐ. ሆቴል መ. የምግብ ሬስቶራንት	ጥር 15, 2021 እስከ ግንቦት 30, 2021
102	የብር አይነት	ሀ. 1 ብር ለ. 5 ብር ሐ. 10 ብር መ. 50 ብር ሠ.100 ብር ረ. 200 ብር	
103	የብር ሁኔታ	ሀ. የድሮ ለ. አዲሱ	
104	የብር አካላዊ ሁኔታ	ሀ. ንጹህ ለ. የቆሸሸ ሐ. በጠም የቆሸሸ	
105	የአንቲባዮቲክስ አይነት	ሀ. erythromycin 5µg ለ. Cefoxitin 30µg ሐ. Gentamicin 10µg መ. penicillin 10IU ሠ. Clindamycin 2µg ረ. Tetracycline 30µg ሰ. Ciprofloxacin 30µg ሸ. Amoxicillin 30µg	

		ቀ. Ampicillin-G 10µg በ. Amikacin 30µg ተ. Ceftazidime 30µg ቸ. Cephalothin 30µg ነ. Cefazolin አ. Trimethoprim + sulfamethoxaz 2.25/23.75µg ከ. Cefepime 30µg ወ. Meropenem 10µg ዐ. Chloramphenicol 30µg	

12. Declaration

I, the undersigned, declare that this M.Sc. thesis is my original work, has not been presented for a degree in this or any other university and that all sources of materials used for the thesis have been duly acknowledged.

M.Sc. candidate: **Hiko Negeho (B.Sc.)**

Signature: _____

Date of submission: _____

Place: Addis Ababa, Ethiopia.

This thesis has been submitted with our approval as advisors.

Approval of the primary Advisor

Name of the primary advisor: Mr. **Kassu Desta (BSc, MSc, Assistant professor, PhD)**

Signature: _____

Date: _____

Place: Addis Ababa, Ethiopia.

Approval of the Co-Advisor

Name of the Co-advisor: Mr. **Melese Hailu (MSc, PhD Fellow)**

Signature: _____

Date: _____

Place: Addis Ababa, Ethiopia.