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Catheter Associated Vancomycin Resistant *Enterococci* (VRE) among patients admitted to Yekatit 12 Hospital Medical College in Addis Ababa, Ethiopia

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ABBREVIATIONS

AMR----------Antimicrobial resistance

ATCC ----------American type culture collection

BAP ----------blood agar plate

CA-UTI----------Catheter associated urinary tract infections

CA-VRE---------- Catheter Associated Vancomycin resistant *Enterococci*

CHS ----------college of health science

CLSI----------Clinical and laboratory standards institute

DDT----------Dilution and disc diffusion test

ESBL ----------Extended spectrum beta-lactamase

HAI---------- healthcare-associated infections

HCW---------- health care worker

ICU----------intensive care unit

IRB---------- Institutional review board

KAP----------Knowledge, attitude and practice

MIC---------- minimum inhibitory concentration

MLST ----------Multi-locus sequence typing

MOSHE----------ministry of science and higher education

MRSA ----------Methicillin resistance *staphylococcus aureus*

NAUTI---------- nosocomial acquired urinary tract infections

NMI----------National metrological institute

PCR----------polymerase chain reaction

PFGE ----------Pulsed-field gel electrophoresis

SPSS ---------- Statistical Package for the Social Sciences

UC---------- Urinary Catheters

UTI---------- Urinary tract infections

VRE----------Vancomycin-resistant *Enterococci*

VSE---------- vancomycin susceptible*Enterococcus*

ABSTRACTS

Background: Catheter associated urinary tract infections (CA-UTIs) are common cause of hospital acquired infections. The emergence of Vancomycin-resistant *Enterococcus* (VRE), poses a significant problem in the management of CA-UTI. There is scarcity of information on the prevalence and drug resistance pattern of VRE among catheterized patients in the current study setting.

Objective: To determine the magnitude & prevalence of **CA-UTI** and **CA-VRE** in catheterized patients and to assess the knowledge and practice of health professionals on CA-UTI.

Methods: A hospital based cross-sectional study was conducted on 270 catheterized patients admitted to Yekatit 12 Hospital Medical College from December 2020 to June 2021. A total of 121 health professionals were also assessed for their KAP on CA-UTI. Urine samples were collected and inoculated on to blood and MacConkey agars, incubated at 35-37°C for 24 hours aerobically. Phenotypic identification was carried out by biochemical tests. Vancomycin resistance for *Entrococcus* was performed by E-test. Data on level of knowledge and practice on CA-UTI was collected from health professionals using questionnaire and analysed by using SPSS version 20.P-value < 0.05 was used as statistically significant.

Result: CA-UTI was detected in 31.9% of the study participants. *E.coli* was the most prevalent isolate (9.3%) followed by *Enterococci* (7.4%).The *Enterococcus* isolates showed highest resistance for penicillin (95%) followed by Doxycycline (55%) and erythromycin(50%). In contrast, lower resistance was documented for Ciprofloxacin (25%), Meropenem (20%) and Vancomycin (20%).The prevalence of CA-VRE among the *Enterococcus* isolates was 20%. Among health professionals participated in the study, 94.2% had knowledge on CA-UTI. Only 34.7% had awareness on CA-VRE and 19.8% had knowledge on bacterial biofilm formation.

Conclusion: The overall prevalence of CA-UTI and CA-VRE was 31.9% and 20% respectively. Medical ward, intermittent catheterization and previous UTI have strong association with CA-UTI, <0.05. Health professionals have little knowledge on VRE and bacterial biofilm formations. Adherence to safety practice and periodic AMR screening in hospital can decrease the prevalence of CA-VRE and improve treatment outcome of hospitalised patients.

Key words: catheter associated vancomycin resistant *Enterococcus*, knowledge and practice,

1. INTRODUCTION

1.1 Background

The Genera *Enterococcus* is facultative anaerobe gram positive coccus capable of surviving in a harsh condition in nature, found in soil, water and plants. Usually colonizes gastrointestinal tract and female genitourinary tracts in both humans and animals as a normal flora. More than 15 species in this genus, (80-90%) in clinical isolates are *Enterococcus faecalis* and the remaining 10-20 % is *Enterococcus faecium* and other species. Though less virulent, can also be potential pathogens responsible for serious hospital acquired infections, causing urinary tract infections (UTIs), wound infection, bacteraemia, neonatal sepsis and rarely meningitis (1, 2).

Nearly 25% of hospitalized patients usually undergo urinary catheterization. Although it's a necessary intervention, indwelling urinary catheters are also a leading cause of nosocomial or health care associated infection. Up to 40 percentile of health care-associated infections, urinary tract infections are the commonest one; 80%, involve catheter-associated urinary tract infections (3, 4).

Recent survey report shows a urinary catheter is the most common indwelling device, with 17.5% of patients in 66 European hospitals and 23.6% in 183 US hospitals having a urinary catheter following admission. Being female, old age, duration of hospital stay, impaired immunity, prolonged catheterisation, underlying disease and conditions (i.e. diabetes, renal disease, and use of systemic antibiotics) and suboptimal aseptic procedural techniques are risk factors of CAUTI (5). Bacterial pathogens causing UTI can enter the bladder during insertion of the catheter, through the catheter lumen, or around the outside of the catheter. The most common infecting organisms are *Escherichia coli*, *Klebsiella* species, *Proteus species*, *Enterococcus species*, *Pseudomonas species*, and *Enterobacter species*. Bacterial pathogens causing UTI are highly resistant to antibiotics; complicating treatment, increase hospital stays and incurs cost (6).

Enterococcus species are the commonest pathogens causing catheter associated urinary tract infections usually resistant to vancomycin and a major problem in hospital environments,

and are highly prevalent in intensive care units particularly patients with underlying diseases. Hospitals in some countries have established VRE screening in high risk areas and isolation of patients to prevent spread of the resistant pathogen. *Enterococcus* species are capable of producing biofilm, bacterial population attached irreversibly on various biotic and abiotic surfaces, usually colonizes medical devices such as urinary catheters (7).

The existence of biofilm facilitates communications among bacterial populations through biochemical signals, called quorum sensing which attributes horizontal transmission of plasmid-associated drug-resistant gene. Biofilms formed by *Enterococcus* species are highly resistant to immune clearance and antibiotic activities. During catheter insertion deposition of fibrinogen will occur due to inflammatory reactions. It becomes a source of nutrition for *Enterococcus* species, enhances formation of bacterial biofilm which confers resistance to phagocytosis (8).

Catheter-associated Vancomycin resistant *Enterococcus* (CA-VRE) become a major threat to public health worldwide and affects patient's clinical outcome. The prevalence of CA-VRE among *Enterococci* differs in different countries including hospital to hospital in the same country. VRE is an important health concern not only because its infections are difficult to treat but also because its clones can spread within hospitals as well as between regions or countries. Urinary tract infection following catheterisation is a major risk for invasion of vancomycin resistant *Enterococcus* (9).

Understanding the risk factors, existence of bacterial isolates, biofilm formation and antimicrobial susceptibility patterns in catheterized patients is very important for proper treatment and management of CAUTIs since these data are lacking in the study area. Most hospitals do not have strict guidelines for the prevention of CAUTI. Training the health care personnel and introducing the prevention of CAUTI as a high priority in hospitals is strongly associated with decreased incidence of CAUTI (5, 9). The present study was planned to determine the prevalence of catheter associated vancomycin resistant *entrococcus* species among admitted patients with inserted catheter and also to assess the knowledge of various health care personnel regarding the risk of catheterisation for CAUTI and measures to prevent it.

1.2 Statement of the problem

Antimicrobial resistance among catheter associated urinary pathogens has been an increasing problem in the last few decades (5). CAUTI is associated with high mortality, increased length of hospital stay, and increased cost of treatment. Catheter-associated urinary tract infection produces substantial morbidity in hospitalized patients including discomfort, fever, malaise and unnecessary antibiotic use, which may become a potential source of antibiotic resistant organisms. Furthermore, the catheterized urinary tract acts as a reservoir for the accumulation and dissemination of drug resistant organisms to other patients (10).

Majority of admitted patients and those who are subjected to medical procedures usually undergo catheterization for purposive intervention. Indwelling urinary catheter is one of the most frequently performed invasive procedures in admitted patients. Some Studies shown as healthcare workers are major source of microorganisms on their hands, cloths, stethoscopes, phones, gowns and other reused items and are frequently contaminated during clinical care from one patient to another in admission wards. Contaminated devices and colonized hospital environments are highly resistant to detergents and antimicrobial agents (8). Formation of biofilm among microorganism is a natural phenomenon during catheterization and causing health care acquired infection. *Enterococcus* is among highly biofilm producing bacteria, persists for prolonged periods on inanimate objects. Biofilm formation begins immediately after catheter insertion, when organisms adhere to the catheter surface; facilitate horizontal transmission of plasmid-associated drug-resistant gene which attributes antimicrobial resistance (10, 11).

Antimicrobial resistance (AMR) is complex and multidimensional range of resistance mechanisms affecting a wide range of infectious agents. The development of AMR is a natural phenomenon in microorganisms and is accelerated by the selective pressure exerted by use and misuse of antimicrobial agents (12).

With the advent of implantable medical devices, patient care quality has improved to a greater extent. Urinary Catheters (UCs) are devices used in almost 15–25% of hospitalized

patients to manage urinary drainage for those who have undergone any surgeries or have problems with mobility. Each hospital follows standard protocols for the insertion, maintenance, and removal of catheters to prevent microbial contamination by maintaining hygiene practices like hand washing, use of sterile gloves, intermittent catheterization and no-touch insertion techniques(13). Despite following all aseptic procedures, the spread of catheter associated-urinary tract infections (CAUTIs) is unavoidable phenomenon in health care settings (10, 13).

The global burden of vancomycin resistance *Enterococcus* as per WHO 2014 survey report shows that 66,000 infections by *Enterococcus* have been reported annually, and 20,000 infections were caused by vancomycin resistance *Enterococcus* species, 1,300 people dies with this organism every year globally (12). The prevalence of clinically isolated VRE was reported in Europe (4%), Asia–Pacific (11.9%), America (35.5%) and Latin America (12.9%). Increasing prevalence of VRE was also reported from few studies conducted in Ethiopia. Meta analysis result showed that an increased prevalence of VRE in Ethiopia from different clinical samples is as follows. Addis Ababa (26.1%) Amhara (15.0%) Oromia (9.0%) and SNNPR (1.9%) (52).

To the best of our knowledge, published data on catheter associated urinary tract infection with vancomycin resistant *Enterococcus* in Ethiopia are either insufficient or underutilized for action. The level of knowledge and practice on catheter associated urinary tract infection and its preventive measures was not well studied. The rationale of the present study was to determine the prevalence of catheter associated vancomycin resistant *Enterococcus* and to assess the awareness of health professionals about catheter associated urinary tract infections and resistance against antimicrobial agents.

1.3 Significance of the study

Many researchers have done and published a lot about the emergence and prevalence of drug resistance in different clinical samples globally. A lot have been conducted on extended spectrum beta-lactamase resistance (ESBL), methicillin resistance *Staphylococcus aureus* (MRSA) and vancomycin resistance *Enterococcus* (VRE) in different clinical and environmental samples. Studies on catheter associated vancomycin resistant *Enterococcus* (CA-VRE) in Ethiopia are few in number. (2, 17, 18, 42, 52).

The prevalence of VRE has to be known since existence of AMR is a natural phenomenon. The awareness and knowledge of health care worker on device related infections and drug resistance has to be known and measured with evidence based data. Findings will be used as an input to take corrective actions and for improvement plans in infection prevention process.

This study will be valuable to indicate the magnitude of the problem and also will be used as baseline data for others.It will be used as input for the health institutions to initiate AMR surveillance program as a routine activity. It will increase awareness of health care workers about device related infections and consequences of drug resistance. It can be a spring board for further similar studies.

2. LITERATURE REVIEW

Different literatures were reviewed to understand the magnitude of the catheter associated urinary tract infections as well as vancomycin drug resistance patterns against *entrococcus* species.

2.1 Catheter associated UTI and vancomycin resistance patterns

A Cross-sectional study to determine Prevalence and antimicrobial susceptibility pattern of *Enterococcus species* isolated from different clinical samples was conducted by Ferede et al. at Black Lion Specialized Hospital, in 2016. The number of participants were 422 with 41.71% females and 58.29% male, 15 *Enterococcus species* were isolated out of 422 and 14 isolates were from hospital admitted patents. All 15 isolates were found as 100 % sensitive to Linezolid and (6.7%) were resistant to Vancomycin. (17).

The second comparative cross-sectional study in Ethiopia was done by Ali, et al, at Dessie Referral Hospital in 2017. From 300 participants, the overall prevalence of *Enterococci* and VRE was 37.3%, and 6.3% respectively. Prevalence of VRE among HIV-positive and negative clients was 5.9% and 7.4%, respectively. Here is a slight difference in VRE prevalence in HIV-positive and negative clients with no justification by the researcher. Antimicrobial susceptibility testing of the isolates was performed on Mueller–Hinton agar (OXOID, UK) by the Kirby–Bauer disk diffusion technique as modified by the Clinical and Laboratory Standard Institute 2017. The statistical analysis on variables with associated factors for Vancomycin Resistant *Enterococci* Shows that Clients with low haemoglobin level were 19.2 times more affected by VRE than normal ones; but no clear explanation for the reason (18).

Another study by Govinda et al, in 2018 to assess the drug resistant pattern and biofilm production in patients with catheter associated urinary tract infection was conducted in Nepal. 105 urine specimens from catheter-associated UTIs suspected patients were processed. Big difference observed in isolates of CAUTI with *E. coli* 56.9% and *Enterococcus* species 1.5%. The greater percentile of antimicrobial resistance was found to

be associated with biofilm producers than biofilm non-producers. The result shows that 46% isolates were biofilm producers. The investigator concluded that High antimicrobial resistance was observed in biofilm producers than non- producers (4).

Lee et al. conducted a study to identify vancomycin-resistant *Enterococci* clones and inter-hospital spread during an outbreak in Taiwan". Rectal samples and infection site specimens were collected from all inpatients in the nephrology ward. VRE strain types were determined by pulsed-field gel electrophoresis (PFGE) and multi-locus sequence typing (MLST). 59 VRE isolates were obtained from 59 patients. The investigator summarized that Increased VRE prevalence is due to cross transmission of VRE clones from undetected VRE carriers and health care workers are the modulator of the process. To avoid cross transmission of VRE in hospital wards, an infection control policy for VRE should include asymptomatic VRE colonization, and thus, active surveillance of VRE during admission; subsequent isolation and appropriate hand-washing practices may be necessary to prevent the spread of VRE within a hospital environment (19). Another study by Alotaibi and Bukhari (20) was conducted in King Saud University. Prospective investigations were carried out on patients with clinically significant *enterococcal* infection attending hospital clinics, emergency rooms, medical and surgical wards, and Intensive Care Units. The clinical significance of the *Enterococcus* isolates was assessed retrospectively by analysing the clinical criteria such as catheterization in UTIs, signs of sepsis, and other laboratory tests such as leucocytosis. A total of 231*Enterococci* were isolated from blood, urine, stool, and other clinical samples. Urine in (41.4%) isolates followed by blood (27.2%) and wound swab (19.4%) were the most commonly involved specimens. VRE were more significant from blood specimens while VSE were significantly more predominant from urine specimens. The most common isolated *Enterococcus* species was *E. faecalis* (73.2%) followed by *E. faecium* (22.9%), and other species were (3.9%). From the total, 40 (17.3%) isolates were VRE. Of these, 33 (62.3%) VRE were *E. faecium* while only 2(1.2%) were *E. faecalis* (20).

Indian researchers, Biswas et al's findings showed us the clinical correlation between the presences of vancomycin-resistant *Enterococcus* genes in colonized patients in clinical isolates. A total of 500 *Enterococci* (250 from clinical samples and 250 from fecal samples

of colonized patients) were processed, out of which 37(7.4%) isolates were vancomycin resistant and 23(4.6%) showed reduced susceptibility to vancomycin by phenotypic agar dilution method. However, by disc diffusion test (DDT), 34(6.8%) strains were found to be resistant to vancomycin and 20(4.0%) strains were found to show reduced susceptibility. Both agar dilution and disc diffusion test (DDT) methods showed a comparable similarity in screening performance with slight differences. The overall prevalence of vancomycin resistant *Enterococci* in colonized patients was about 9.6%. Prior administration of antibiotics had significant effect on VRE carriage. Urinary tract infection was the most common infection caused by vancomycin susceptible *Enterococci* (49.0%) and VRE, (36.1%). There was no significant difference in the distribution of VRE and VSE in different infection types (21).

A study conducted in Serbia by Milan & Ivan to assess catheter-associated nosocomial acquired urinary tract infections and influence of antibiotic resistance on antimicrobial therapy was explained in detail. 589 hospitalized patients for operative treatment in the Urology clinic were studied. The study focuses to address the need for frequent re-evaluation of the prevalence of pathogens and resistance profile in CAUTI. Patients with NAUTI as well as patients with catheter associated urinary tract infections (CAUTI) have a similar resistance and similar microorganisms isolated as causative agents. As outlined by the study, urinary catheter is the most important predisposing factor for both CAUTI and spread of bacterial resistance. Isolated pathogens of CAUTI are *E. coli* (65%), *Proteus mirabilis* (26%), and *Enterococcus* (9%). From gram positive organisms *Enterococcus* was the predominant as per the study findings (22).

J.W. Warren (8) from University of Maryland, USA, has conducted a study to investigate the prevalence of “Catheter-associated urinary tract infections” acquired in both hospitals and nursing homes associated with urinary catheterization. Insertion of a catheter may carry urethral organisms into the bladder. Most bacterial strains that enter the catheterized urinary tract are able to multiply to high concentrations within a day. Biofilm, which covers and secures bacteria against a catheter or mucosal surface, has been demonstrated on drainage bags, catheters, and the uroepithelium. Organisms contained within the biofilm appear to be well-protected from hosts defences, and antibiotics.

A study explains about the effect of short term and long term catheterisation with regard to Catheter-associated urinary tract infections and bacterial profile. In short term catheterization, common bacterial species are *Escherichia coli, P. aeruginosa, K. pneumoniae, P. mirabilis, S. epidermidis, Enterococcus* species.Most bacterial isolates in short-term catheterization causes asymptomatic CAUTI. In contrast, microorganisms usually isolated from long-term catheterized patients are 95% polymicrobial type, commonly two or more bacterial species with concentrations of 10^5cfu/ml or more. The study recommends instead of urethral catheterization, suprapubic catheterization have shown significant benefits in terms of lowering the incidences of Catheter-associated urinary tract infection in hospitalised patients. Urinary tract infections are the commonest healthcare-associated infections (HAI), accounts up to 40%. The risk of acquiring a catheter-associated infection increases with the duration of catheterisation, the daily rate is 5%, by 4 weeks almost 100% of patients may develop healthcare-associated infections. To minimize the risk, urinary catheters must only be inserted when clear medical indications observed and should be removed as soon as no longer needed. Under normal circumstances urethral flora, migrating into bladder, are constantly flushed out during urination. When a catheter is inserted, flushing mechanism will be circumvented and perineal and urethral flora can pass up into the bladder in the fluid layer between the outside of the catheter and the urethral mucosa or in the catheter lumen (i.e., endogenous). Because of this, bladder colonisation is inevitable if catheters are left inserted for prolonged periods. Bladder infection can also be caused by bacterial reflux back flow from contaminated urine in the drainage bag. Closed drainage systems reduce onset of infection by limiting access of bacteria to the urine. The study evidences that hands of health care workers are also potential source to contaminate the urinary catheter during insertion or management (i.e., exogenous), and microorganisms causing healthcare-associated UTIs in hospitalised patients are usually resistant to antibiotics (8).

Fatholahzadeh et al, have done surveillance from three different hospitals to detect vancomycin resistant *Enterococci* (VRE) isolated from urinary tract infections in Iran in 2006. From 120 *Enterococci* isolates 7% were resistant to vancomycin. Various VRE species were isolated, including *E. faecalis* 38% *E. faecium* 25%, *E. mundtii* 25%, and *E.*

raffinosus 13%. Majority (75%) of VRE from ACMC H/L with the highest diversity. In contrast, 25% of VRE and only *E. faecium* strains was isolated from Mehrad hospital. Interestingly, all Enterococci isolates from Pars hospital were sensitive to vancomycin and no VRE was recovered.

The susceptibility testing was performed by disk agar diffusion method and minimum inhibitory concentration (MIC) value for each VRE isolate was determined by the agar dilution method and the vanA gene was detected by PCR. (23).

Another study was conducted by Puri et al, to see incidence of infection following short term vs long-term urethral catheterization with regard to antibiotic resistance pattern in catheterized patients who had indwelling catheters for >48 h, in India. The finding shows that, 68 (8.5%) adult inpatients acquired urinary tract infection following indwelling catheterizations with significantly higher risks for female, elderly patients, critically ill and patients on prolonged catheterization. Among the pathogens, *E. coli* was the commonest isolate (32.9%) followed by Pseudomonas sp. (15.1%), *S. aureus* (12.3%) and Candida sp. (13.7%). all G +Ve pathogens including *Enterococcus* were sensitive to Vancomycin (24).

According to Bagchi, et al's study, the urinary catheter is an essential device in modern medicine, used to relieve obstructions, to drain urine for comatose or sedated patients, and to measure urinary output in severely ill patients. Unfortunately, when used inappropriately or left too long, it becomes hazard to the patients instead of intended purpose. As explain in detail, urinary tract infections are the predominant healthcare-associated disease caused by instrumentation of the urinary tract, accounting for more than 30% of infections in hospitals. Urine samples from all patients admitted without history of UTI and indwelling catheter was collected aseptically, analysed by microscopy and cultured for microbiological profiling. Antibiogram was studied and CAUTI was observed in 64 out of 220 samples with most frequent *E.coli* (34.85%), Klebsiella (19.7%), Pseudomonas (12.12%), Candida (10.6%), *Enterococcus* (6.06%), CONS (6.06%), S. aureus (4.55%), Citrobacter (3.03%) Proteus (3.03%). Infections were more with female sex, prolonged catheterization, old age and diabetes. Longer duration of catheterization increased the chances of CAUTI (25).

Jayakumar S et al, has conducted a Prospective study on Short Term Urinary Catheterized Patients in Intensive Care Unit (ICU) in India, Mahsa University College to assess the magnitude and burden of Catheter-Associated Urinary Tract Infection (CAUTI), he found that up to 40% of all nosocomial infections are associated indwelling urinary catheter comprises a huge reservoir of resistant pathogens in hospitals particularly in critical care units. 100 urine samples were collected catheterized patients in a hospital Intensive Care Unit (ICU). Phenotypic identification was performed following antibiotic sensitivity testing using Kirby-Bauer disc diffusion method.

Out of 100, 32 showed growth on 5th day and remaining 68 were culture negative up to 7th day. None of the culture positive samples had pus cells. E. coli (35%) was the predominant isolate followed by *P.aeruginosa, K. pneumoniae, E. faecium*, Candida spp. (15% each) and Coagulase negative Staphylococcus (5%). Among the Gram negative organisms, E. coli and *K. pneumonia*e showed Extended Spectrum Beta Lactamases (ESBL) producers. In conclusion catheterized asymptomatic patient are a major reservoir of Multidrug resistant pathogens in hospitals and periodic surveillance for nosocomial infections in hospital is a very good protocol to control the spread of nosocomial infections (26).

A cross sectional study by Kalpana Devi Venkatesan et al, in India was done to determine the emergence of *Enterococci* as uropathogens and their antimicrobial susceptibility pattern revealed that 175 *Enterococcus* spp were isolated from patients with urinary tract infection. Identification of the *Enterococcal* isolates was done using conventional method. Antibiotic susceptibility testing was performed by modified Kirby-Bauer disk diffusion method as per Clinical Laboratory Standard Institute (CLSI) guidelines. *E. faecalis* (77.7%) was the predominant species isolated followed by *E. faecium* (20%) and *E. durans* (2.3%).The sensitivity pattern showed an increased resistance to antibiotics like erythromycin (66.3%), ciprofloxacin (56%) and penicillin (49.7%). Among the isolated Enterococci, 5.7% were vancomycin resistant and all the rest were sensitive to linezolid. Based on their conclusion, Enterococci have emerged from being harmless commensals to versatile lethal pathogens and *E. faecalis* was the most frequently isolated species followed *E. faecium* (27).

Agrawal Neha and his colleagues (29) have done a hospital based cross sectional study on antimicrobial susceptibility pattern of *Enterococcus* species isolated from urine samples in India. 105 *Enterococcus* spp were identified by using gram staining, catalase test, esculin hydrolysis test, and (PYR test). Antimicrobial susceptibility testing was done by Kirby Bauer disc diffusion Method as per CLSI guideline. The result he found shows, *Enterococcal* isolates were more from in-patients (76%) than outpatients (24%). Among in-patients (ward/ICU) isolates, 46.24% of the isolates were from catheterized patients. Among the *Enterococci* isolates (51.42%) were from females and (48.57%) were from males. E. faecium was predominant (67.62%) over *E. faecalis* (32.38%). Isolated *Enterococci* showed highest resistance to fluoroquinolones (88% to 94%) and least resistance to linezolid (6.66%). Vancomycin resistant *Enterococci* (VRE) were found in 16.19% of cases while 62.85% Enterococci isolates were high level gentamicin resistant.

Based on the findings, Enterococci is emerging as a common isolates of vancomycin resistance, from urine sample and a serious challenge for physicians treating patients with infection due to these microorganisms in hospitalized patients (28).

Microbial flora in patients with indwelling catheter was studied by Manish N. et al, to determine the microbiological profile and the sensitivity pattern of Catheter Associated Urinary Tract Infections in India. 100 patients with indwelling Foley's catheter were enrolled in the study in medical and surgical wards. The findings showed that the most common organism colonizing and causing catheter associated urinary tract infection found to be *E.coli* (57%) followed by Klebsiella Sp.(20%), Staphylococcus (8%), *Enterococcus* Sp. (6%), *Pseudomonas aeruginosa* (5%) and Acinetobacter Sp. (4%) (29).

Indian researchers Naveen, Nagraj and Latha, have conducted a study on Catheter associated urinary tract infections (CAUTI). The result revealed that indwelling urinary catheter becomes a potential source for drug resistance microorganisms. 126 urine samples from catheterized patients were collected and processed microbiologically and antimicrobial sensitivity was performed. 26 developed CAUTI. *E. coli* (65.3%) was predominant followed by Proteus (15.38%), *Klebsiella* (7.6%). Among gram positives, *Enterococcus* spp. was (7.6%) followed by *S. epidermidis* (3.8%) (30).

2.2 Awareness of health professionals on CA-UTI

Knowledge, awareness and practice of health care professionals were surveyed by using a self-administered questioner adopted from WHO COVID-19 risk assessment and management protocol. The purpose of the survey was to assess the level of health care professionals on catheter associated urinary tract infection due to indwelling catheter and preventive measures practiced.

Pandian et al, conducted a study to explore the knowledge, attitude, and practice levels and prevention of CAUTI among the health care professionals. The result showed that 28.4% and 71.6% of the participants had moderate and adequate knowledge respectively about CAUTI. 4.2% and 95.8% of respondents had moderate and adequate practice towards the prevention of CAUTI. They also found that the experience of participants showed statistically significant association with the level of practice. The researcher recommends that medical education programs, frequent auditing with a checklist about catheter care must be conducted to maintain the knowledge and practice of health care professionals (31)

Nigerian researchers Emelda et al, have shown the level of knowledge on antimicrobial resistance and drug prescribing behaviour of health care professionals. the finding explained as (50.3%) of participants agreed that their prescribing behaviour could promote antimicrobial resistance. 49.2% had a good knowledge of AMR and physicians had significantly better knowledge than other HCWs ($X2 = 69.59$, $P<0.001$). Over 60.3% admitted prescribing antibiotics just to be on the safe side (32).

The level of Knowledge and attitude of doctors and nurses regarding prevention of catheter-associated urinary tract infection was studied by Manisha et al. The findings revealed that doctors had better knowledge than nurses regarding the preventive measures for CAUTI. Health care professionals posted in high-risk areas such as ICUs had significantly better knowledge than posted in wards regarding various indications. Experience had no effect on the knowledge regarding indication for catheterization and preventive measures. More than 90% of the health care professionals also felt that education regarding basic catheter care would also help prevent CAUTI (33).

2.3 Hypothesis

The prevalence of vancomycin resistant *Enterococcus* species among catheterised clients at Yekatit 12 hospital medical college Addis Ababa, Will be quite different from studies done in elsewhere other than Addis Ababa,

3. OBJECTIVE

3.1 General objectives

The general objective of this study was to determine the prevalence of vancomycin resistant *Enterococcus* and catheter associated urinary tract infections among catheterised clients and to assess the knowledge, awareness and practice of health professionals on catheter associated urinary tract infections by drug resistance organisms in Yekatit 12 hospital medical college, Addis Ababa, Ethiopia, 2020.

3.2 Specific objectives

I. To determine the prevalence of catheter associated urinary tract infections among catheterised patients at Yekatit 12 hospital medical college.

II. To determine the prevalence of catheter associated vancomycin resistance *Enterococcus* species at Yekatit 12 hospital medical college.

III. To assess the knowledge and practice of health care workers on catheter associated drug resistant organisms inYekatit 12 hospital medical college.

4. MATERIALS AND METHODS

4.1 Study area

The study was conducted in Yekatit 12 hospital medical college which is located at north of Addis Ababa, proximate to Addis Ababa University main campus 6 kilo. It is one of the hospitals under Addis Ababa City Administration Health Bureau that has been giving routine health services for residents of Addis Ababa city and receives referral cases from different regional states of Ethiopia. It has 32 departments providing all major and minor clinical service with 375 admission beds, 22 referral OPD and clinics. The unique feature of the hospital medical college is providing plastic and reconstructive surgery and the only center for speech therapy treatment in Ethiopia

In 2011 G.C the hospital upgraded to medical college providing undergraduate and postgraduate education programs in different disciplines. Five rounds of medical Doctors in medicine have been graduated to date.

The college has microbiology laboratory performing culture and sensitivity tests for routine clinical samples from the hospital clients and are staffed with well experienced and dedicated microbiologists.

4.2 Study Design

Hospital based cross sectional design was applied to determine the prevalence of catheter associated vancomycin resist ant *Enterococcus* among catheterised clients and to assess the knowledge and practice of health professionals about catheter associated urinary tract infection by drug resistance organisms at Yekatit 12 hospital medical college, Addis Ababa, Ethiopia

4.3 Study Period.

The data and sample collection period was from December, 2020 to June, 2021G.C. The prolonged sample collection period was because of Covid-19 pandemic, the hospital's functionality was limited to provide only emergency service and admission wards were occupied with few patients.

4.4 Population

4.4.1 Source population

- For objective 1 & 2: All admitted patients to Yekatit 12 hospital medical college in Addis Ababa, Ethiopia were source population for this study.
- For Objective 3: All health care workers at Yekatit 12 hospital medical college

4.4.2 Study population

- For Objective 1 and 2: catheterized patients admitted to Yekatit 12 hospital medical college during the study period
- For Objective 3: Healthcare professionals who were assigned in the admission and emergency wards during the sample collection time.

4.5 Inclusion and exclusion criteria

4.5.1 Inclusion criteria

- All admitted patients above 18 years old with indwelling catheter irrespective of gender were included in the study.
- All health care professionals who were assigned in the admission and emergency ward during the sample collection time were included in the study.

4.5.2 Exclusion criteria

- Admitted patients without urinary catheter and those who are stayed with inserted catheter for prolonged period (more than 30 days) were not included in the study since the maximum hospital stay was not more than a month.
- Patients and health care workers who were not willing to participate in the study.
- Students who attaches for clinical practice in the hospital were not included in the study.

4.6 Study Variables

4.6.1 Dependent variables

- Catheter associated vancomycin resistant *Enterococcus* species
- Catheter associated urinary tract infection
- Health Professionals' awareness on catheter associated UTI and drug resistance

4.6.2 Independent variables

- Sex
- age
- Educational status
- Work experience
- Marital status
- Profession
- Admission ward place
- Duration of admission
- Underline disease/ co morbidity
- Previous UTI and catheterisation
- Infection prevention training

4.6.3 Sample Size determination

A single population proportion formula was used for the determination of the sample size by considering the following assumptions: prevalence of VRE 22.7% from a study done in Jimma by Toru et al (2), level of significance = 0.05, at 95% confidence interval, therefore,

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

$$n = \frac{(1.96)^2\ 0.227(1-0.227)}{(0.5)^2}$$

n= 270

4.7 Sampling Procedure Data collection

4.7.1 Data collection

A convenient sampling technique was applied to collect the sample and data. The importance of the study was explained for the participants. The patient's necessary medical data such as socio-demography, co-morbidities, reasons for admission, admission ward place, duration of hospitalization and duration of catheterization was obtained from patients' medical records supplemented by patient face to face interview. For health care workers knowledge assessment, structured and self-administered questionnaire was adopted from the WHO Risk assessment Interim guideline 2020 (35). Written consent form was provided and signed by the study participants before sampling.

4.7.2 Sampling Procedure

Sample collectors were given training on how to collect urine sample from catheterised patients aseptically. Sterile urine sample collecting cups were provided to the sample collector. Urine specimens were obtained by using aseptic technique (disinfecting with 70% alcohol) by puncturing the catheter bag with a syringe. Catheterised urine was taken in this way because risk of contamination. Samples from freshly removed catheter, the catheter tips were immersed in to sample cup containing sterile 0.85N saline.

The sample transport, storage and sample quality was supervised by the principal investigator and transported immediately to bacteriology teaching laboratory for processing. The leftover urine samples were stored for 24 hours in the refrigerator to prevent secondary sampling due to power interruption or other unavoidable circumstances in the laboratory. The awareness assessment questionnaires were administered for all health care workers working in admission wards and emergency during sample collection and administered back after filled all the necessary information. The questionnaire has to parts describing socio-demographic details and awareness on catheter associated UTI on admitted patients.

4.8 Sample processing and pathogen identification

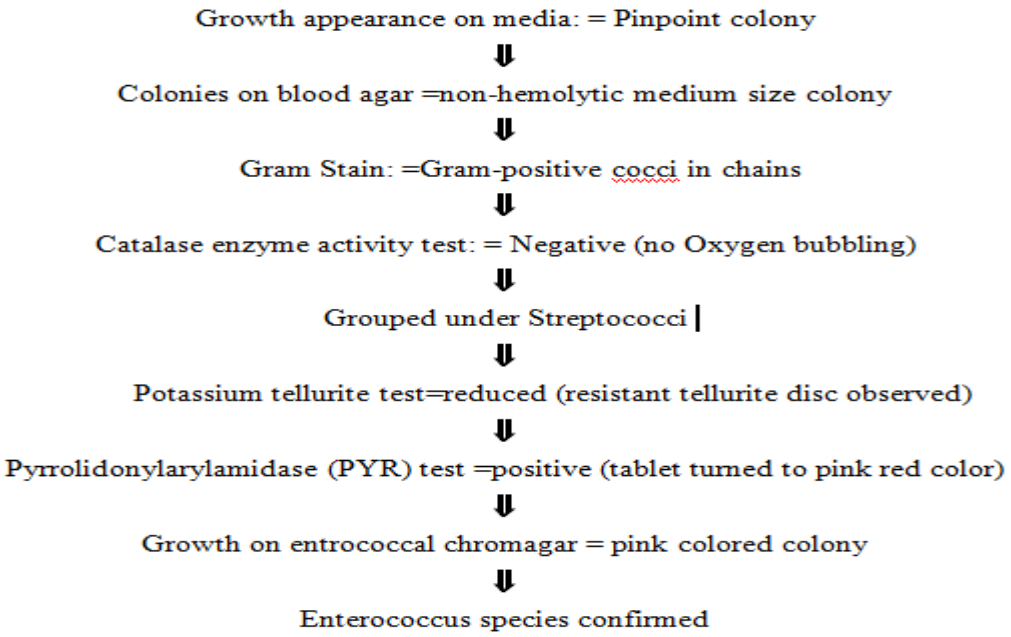
Collected Urine sample were inoculated simultaneously on to primary culture media; i.e. blood agar plate (BAP) and MacConkey agar plate [Oxoid Ltd Co, UK] by using 1µl calibrated wire loop aseptically. The sterility, PH, and growth performance evaluation test or internal quality control for each culture media was performed by using American type culture collection (ATCC) strains based on the clinical laboratory standard institute (CLSI-2019, M100) guideline. Inoculated media plates were incubated in to the temperature adjusted to 35-37**ºC** incubator for 18 to 24 hours. The temperature of the incubator was checked and recorded daily with calibrated thermometer.

Routine manual conventional bacterial biochemical identification methods were applied for the bacterial isolation process. Incubated media plates were observed visually and bacterial growth on primary culture media was evaluated based on colony characteristic. Haemolysis reaction under or around the bacterial colony on blood agar was observed and evaluated as

beta, alpha or non-haemolytic reaction protocols. Number of bacterial colonies was counted and growth of bacteria 10^4-10^5CFU was considered as significant since the samples were collected from catheterised clients. It is believed that catheterised urine is sterile and free of organisms. Growth of organism more than two types, the predominant and commonest known aetiology was considered as significant bacteria.

For gram positive organisms, Gram stain, catalase test, coagulase test, and Pyrrolidonylarylamidase (PYR), tests and other identification tests were utilised to isolate *Enterococcus* species from other organisms. Sub-culturing was performed on chrome agar (Chrome ID (C-ID) medium (bioMerieux, France) to confirm isolates of *Enterococcus* species. For gram-negative bacteria: Oxidase, carbohydrate utilization test, indole test, mannitol fermentation test, decarboxylation, citrate utilization, motility test, and urea utilization test was performed to identify each individual organisms and interpreted as per laboratory SOPs and job aids. Figure-1.

Figure 1: Enterococcus Species Identification job aid (n = 270) with urinary catheter attending Yekatit 12 Hospital medical college, Addis Ababa, Ethiopia, 2021



Adopted from Ellen jo baron job aid, ASM 2012

4.9 Antimicrobial susceptibility testing

Antibiotic susceptibility testing was carried out by the Kirby-Bauer disc diffusion method on Muller-Hinton agar according to the CLSI guideline (36). The turbidity of the prepared inoculums suspension was adjusted to 0.5 McFarland's standard with densitometer. Thickness of Muller-Hinton agar was measured to fit 4mm+/- 0.2 to prevent false resistance because of vertical diffusion. Antimicrobial susceptibility tests against Ceftriaxone 30µg, Doxycycline 30µg, Erythromycin 15µg, meropenem 30µg, ciprofloxacin 5µgVancomycin 30µg, and Penicillin 10 units were performed for *Enterococcus*. E-test was utilised as a confirmatory check for vancomycin resistant *Enterococcus* with MIC value of 0.016—256 µg concentration gradient to see the difference in drug resistance pattern.

4.10 Operational Definitions

UTI is defined as the presence of symptoms and signs compatible with UTI and a urine culture growing $\geq 10^5$ CFU/ml with ≥1 bacterial species.

CA-UTI is defined as a UTI occurring in a person with a UC in place or in whom a catheter was removed within the previous 48 hours (49).

CA-VRE is a strain of bacteria that can cause urinary tract infection. Vancomycin is used to kill the bacteria. However, VRE is resistant to vancomycin and makes it difficult to treat. VRE most commonly causes an infection in the urinary tract, can easily spread from person to person in hospitals (51)

4.11 Internal Quality assurance for antimicrobials

All preparations of antibiotic performance efficacy were tested for ATCC strains of *Enterococcus, E. Faecalis* ATCC 29212. Results fall within the Quality Control ranges as stated in CLSI document M100 -2019, for all isolates were accepted. For those antibiotics where a failed QC was recorded, all results were discarded for that test and the testing for that isolate was repeated (17).

4.12 Data quality Assurance

10 % of the questionnaire for the health care worker was pre-tested in two hospitalsout of the study site to ensure that questionnaire was clear for respondents. After pre-testing, some modification was made for unclear and difficult questions. These pre-test data was not included in the analysis of the study. The completeness, cleanness and clerical errors were rechecked by the PI every day.

4.13 Data processing, management and interpretation

All the recorded data was entered manually in to computer for analysis. Statistical analysis was done using SPSS v.23. Appropriate descriptive and inferential statistics was calculated. For all data analysis level of significance was set at 5% α value and 95% confidence intervals. Results with P value <0.05 was considered as statistically significant.

4.14 Ethical consideration

Ethical approval was obtained from Departmental ethics committee of Addis Ababa University, department of Microbiology, Parasitology and Immunology. Permission letter to collect specimens was obtained from Yekatit 12 hospital medical college research and publication office. Clinically important information was communicated with the attending clinical staffs for benefits of the patient. All the records of patient's data was stored in secured lockable drawer and kept confidential by principal investigator.

4.15 Dissemination of Results

The results of the study will be submitted to Addis Ababa University, College of Health Sciences, Department of Microbiology, Parasitology and Immunology and Yekatit 12 Hospital medical college. In addition, it will be presented in annual conferences of professional societies and other concerned bodies. The finding of the study will be also presented to the medical scientific community and manuscript will be submitted to peer-reviewed journals for publication.

5. RESULT

A total of 270 study participants with indwelling urinary catheter admitted to different wards and 121 health professionals working in several wards of the Yekatit 12 hospital medical college were enrolled in this study.

5.1 Socio-demographic characteristics of participants with indwelling urinary catheter

Majority of the study participants with indwelling urinary catheter were from 31-50 years old 88 (26.2%) followed by 51-70 years 82 (30.4%), >70 years 70 (25.9%) and 18-30 years 30 (11.1%). Regarding sex, 141(52.2%) were males. Majority of the participants were from medical admission ward 182(67.8%) followed by surgical ward 56 (20.7%), and emergency 32 (11.9 %).

Most of the study participants 167 (61.9%) had a hospital stay of 4-10 days followed by 1-3 days 64 (23.7%) and ≥10 days 39 (14.4%). Of the study participants, 122(45.2%) had underline disease; 78(28.9%) had hypertension, 32 (11.9 %) had Diabetes mellitus, and 12(4.4%) had HIV, STI and cancer.

About half of the participants 136 (50.4%) have stayed with inserted catheter for 4-9 days, 78(28.9%) for 1-3 days, 29(10.7%) for 10-30 days and 27(10%) had intermittent catheter insertion. 44 (16.3%) participants had history of previous UTI and 46 (17%) had history of previous catheterization.

At the time of sample collection 217(80.4%) of participants were on different antibiotics and 130(48.1%) participants were married 85(31.5%) and 55(20.4%) participants were single and divorced respectively.

91(33.7%) participants were illiterate, 129 (47.8%) were 10/12 grade completed and 44 (16.3%) of the participants were diploma and above in their educational status.

5.2 Bacterial Profile from participants with urinary catheter

Out of 270 catheterized patients, bacterial CAUTIs was identified in 86 patients giving a prevalence rate of 31.9%. CAUTI caused by *Enterococcus* species was 20(7.4%).

Among the bacterial species, *E.coli* 25 (9.3%) was the most predominant isolate followed by *Enterococcus* species 20 (7.4%), *Klebsiella* species 19 (7%).

More than two third of the isolates were gram negative bacteria 60/86, (69.8%) whereas gram positive bacteria constituted 26/86, (30.2%). **Table-1**

Table 1: Bacterial profile from study participants (n = 270) with urinary catheter attending Yekatit 12 Hospital medical college, Addis Ababa, Ethiopia, 2021

Name of Isolate	Frequency	Percentage (%)
No bacterial growth detected	**184**	**68.1**
Escherichia coli	**25**	**9.3**
Enterococcus species	**20**	**7.4**
Klebsiella species	**19**	**7**
Proteus species	**4**	**1.5**
Pseudomonas species	**4**	**1.5**
Coagulase –ve staphylococci	**3**	**1.1**
Staphylococcus aureus	**3**	**1.1**
***Citrobacter* species**	**3**	**1.1**
***Acinetobacter* species**	**2**	**0.7**
***Enterobacter* species**	**2**	**0.7**
***Salmonella* species**	**1**	**0.4**

5.3 Factors associated with bacterial CA-UTI

To see whether there was significant association between dependent and independent Variables, bivariate and multivariate logistic regression were performed. Significant association was observed in marital status, place of admission ward, history of Previous UTI and duration of current catheterization. Being unmarried was found to be 55% less likely vulnerable to acquire CAUTI than married and divorced clients with P value of 0.045. Patients admitted to medical ward and emergency are twice and three times more likely to have CAUTI than those in surgical wards with p value of 0.022(1.244-8.426) and 0.016(1.17-7.45), respectively. **Table -2**

Table 2: Variables associated with UTI of study participants with urinary catheter (n =270) attending at Yekatit 12 Hospital medical college, Addis Ababa, Ethiopia, 2021

Categorical Variables	CA-UTI		P-value	COR [95 % CL]	P-value	AOR [95 % CL]
	Yes n (%)	No n (%)				
Age						
18-30 Years	12(4.4)	18(6.7)		1		
31-50 Years	34(12.6)	54(20)	.057	094(041-2.2)	.340	
51-70 Years	45(16.7)	37(13.7)	.487	1.82(0.78-4.27)	.299	
>71 Years	36(13.3)	34(12.6)	.340	1.6(0.67-3.78)	.640	
sex						
Male	66(24.4)	75(27.8)		1		
Female	61(22.6)	68(25.2)	.809	1.02(0.63-1.64)	.809	
Marital status						
Married	72(26.7)	58(21.5)	.505	1.2(.63-2.25)	.506	1.21(0.61-2.39)
single	27(10)	58(21.5)	.003	0.45(.22-.90)*	.036	0.45(0.22-0.96)*
divorced	28(10.3)	27(10)		1		1
Education						
Illiterate	60(22.2)	69(25.6)	.939	0.93(0.42-2.1)	.939	
10/12 complete	41(15.2)	50(18.5)	.979	0.88(0.38-2.03)	.979	
Certificate &Diploma	12(4.4)	9(3.3)	.208	1.4(0.46-4.42)	.208	
Degree & above	14(5.2)	15(5.6)		1		
Admission ward						
surgical	22(8.1)	41(15.2)		1		1
medical	88(32.6)	87(32.2)	.008	1.89(1.03-3.84)*	0.016	2.02(1.06,3.83)*
Emergency	17(6.3)	15(5.6)	.045	2.11(0.89-5.03)	0.011	2.9(1.17-7.45)*
Co-morbidity						
Yes	64(23.7)	58(21.5)	.018	1.5(.9-2.4)*	.893	1.040(.584,1.855)
No	63(23.3)	85(31.5)		1		
Previous UTI						
Yes	27(10)	17(6.3)	.002	2(1.03-3.88)*	.040	1.86(0.92-3.76)*
No	100(37)	126(46.7)		1		
Duration of catheterization						
1-3 day	30(11.1)	48(17.8)		1		
4-9 days	61(22.6)	75(27.8)	.344	1.3(0.7-2.29)	.255	1.1(0.61,2.01)
10-20 days	17(6.3)	12(4.4)	.022	2.27(0.95-5.4)	0.540	2.26(0.89,5.69)
Intermittent insertion	19(7)	8(3)	0.033	3.8(1.48-9.76)*	0.005	4.2(1.57,11.22)*

NB. *=p value <0.05

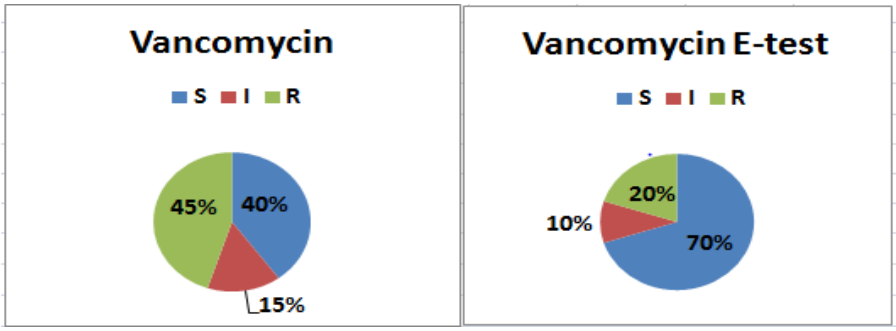
Patients with history of previous UTI are twice more likely subjected to be exposed for CAUTI than those without history of pervious UTI, p value of 0.002 with CL (0.92-3.76). Also significant association was observed in duration of catheterisations. Those who had an intermittent catheter insertion are four times more likely to have CAUTI than short term and medium term catheter duration, p-vale of 0.033 with CL (1.57-11.22). Similar study done in Indonesia showed that Patients aged > 50 years old (P < 0.03) and duration of catheter use (P < 0.03) were both significantly associated with increased risk of developing UTI (41).

No significant association was observed in age, sex educational status, underline disease and duration of hospital admission. During bacterial identification, staphylococci other than *S.Saprophyticus* were considered as contaminant skin flora and yeast cells were also not included in the prevalence rate in catheter associated urinary tract infection. **Table -2**

Table 3: Antimicrobial susceptibility pattern of bacterial isolates from study participants with urinary catheter inYekatit12 hospital medical college, Addis Ababa, Ethiopia, 2021

Antibiotic discs	Susceptibility pattern	*Enterococcus* (n=20)	AST pattern in Percentages %
Penicillin, 10 IU	Sensitive	1	5
	Intermediate	-	-
	Resistant	19	95
Erythromycin, 15µg	Sensitive	8	40
	Intermediate	2	10
	Resistant	10	50
Doxycycline, 30µg	Sensitive	8	40
	Intermediate	1	5
	Resistant	11	55
Ciprofloxacin, 5µg	Sensitive	13	65
	Intermediate	2	10
	Resistant	5	25
Meropenem, 10µg	Sensitive	14	70
	Intermediate	2	10
	Resistant	4	20
Ceftriaxone, 30µg,	Sensitive	10	50
	Intermediate	2	10
	Resistant	8	40
Vancomycin Disk 30µg	Sensitive	8	40
	Intermediate	3	15
	Resistant	9	45
E-Vancomycin 0.016—256 µg	Sensitive	14	70
	Intermediate	2	10
	Resistant	4	20

Figure 2: VRE in disc diffusion and E-test methods at Yekatit 12 Hospital medical college, Addis Ababa, Ethiopia, 2021



Increased prevalence in drug resistance was observed in many classes of drugs. The highest drug resistance rate was observed in penicillin (95%) followed by Doxycycline (55%), erythromycin (50 %); Vancomycin disc diffusion (45%) ceftriaxone (40%) showed greater than 40% of resistance rate. In contrast, meropenem (20%) and ciprofloxacin (25%) were less resistant with the rate below 30%. Huge discrepancy observed in between vancomycin disc diffusion test and E-test with susceptibility rate. For E-Test 70% and disc diffusion test 40% was observed in the current study. Resistance rate was measured as 45% for disc diffusion and 20% for E-test. **Figure-2**

5.4 Awareness assessment of health professionals and associated factors on CAUTI

Awareness assessment on catheter associated urinary tract infection among health professionals working in admission wards during sample collection period was also assessed by using questionnaires. A total of 121 health professionals' were participated in the study with a response rate of 100%. Majority 114(94.2% of participants had knowledge on CAUTI and 97(80.2%) participants have knowledge on existence of multidrug resistance in hospital environments. Only 42(34.7%) participants were aware of vancomycin resistant *entrococcus* causing UTI and only 24(19.8%) had knowledge on bacterial Biofilm formation. Among participants 91(75.2%) said that there are constraints of supply for infection prevention materials. even though the scarcity of consumables safety materials, 88(72.7%) of health care providers adheres in infection control measures to maintain patients safety. Availability of safety guideline was assured by 58(47.9%) of

participants and 62 (51.2%) had training on infection prevention and safety. Of the participants, 78 (64.5%) wash their hands before and after catheter insertion. **Figure 3**

Figure 3: awareness of health professionals (n = 121) at Yekatit 12 hospital medical college, Addis Ababa, Ethiopia, 2021

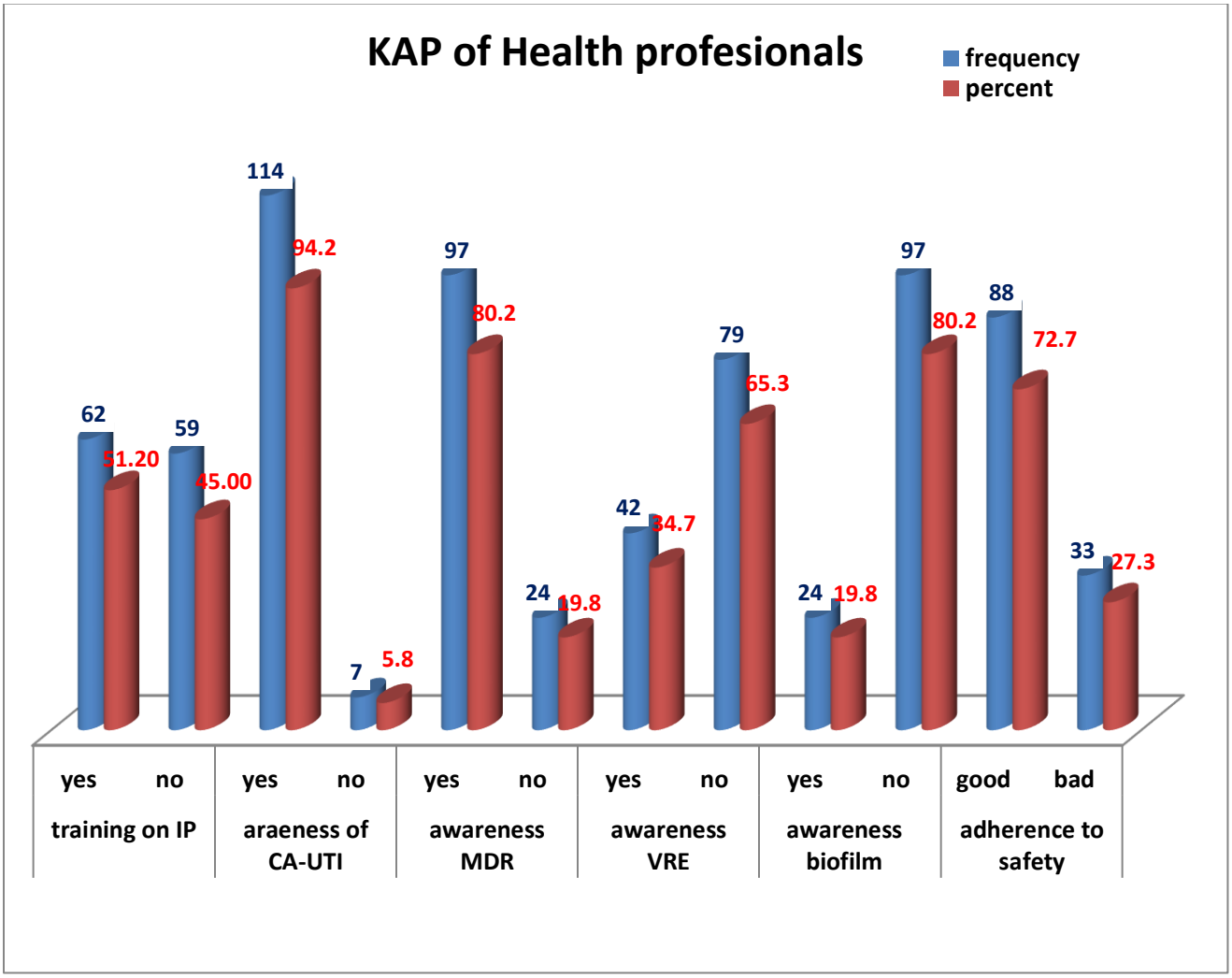


Table 4*:* Socio-demographic Characteristics of health professionals (n = 121) at Yekatit 12 hospital medical college, Addis Ababa, Ethiopia, 2021

Categorical Variables		Frequency	Percentage (%)
age	20-30yrs	16	13
	31-45yrs	92	76
	46-56yrs	7	5.8
	>56yrs	6	4.9
Sex	Male	64	53
	Female	57	47
marital status	married	35	28.8
	Single	75	62
	divorced	11	9.2
work experience	<1 yr	37	30.6
	1-5 yrs	45	37.2
	6-15 yrs	28	23.2
	>16 yrs	11	9.1
Educational status	nurse	72	59.5
	health officer	7	5.8
	doctor	40	33.1
	other	2	1.7

6. DISCUSSION

6.1 Prevalence of CA-UTIs and major isolates

Understanding the existence, determining the prevalence of catheter associated UTI and drug resistance pattern is an important step to formulate an intervention to control emergence and transmission of resistant pathogens. In recent years, an increase in catheter associated UTI with VRE infections have been reported elsewhere worldwide (12).

The current study, documented an overall 31.9% prevalence of catheter associated urinary tract infection due to bacteria. The finding is comparable to studies done in India 29% (25), and Michigan University in USA 31.5% (35). However, the prevalence of CAUTIs in our study is higher than studies done in India 22.4% (40), Egypt 17.5%, and 15.7% (42, 44), and Arbaminch in Ethiopia, 16.88% (33). The current finding is lower than studies conducted in India 40% (26), Egypt 47.8 % (41) and Indonesia 44.4% (43). The difference in prevalence of bacterial CA-UTIs could be due to gender of the participant, implementation of infection prevention practice, catheter insertion care, duration of catheterization and previous catheterisation (33)

A variety of gram positive and gram negative bacteria are implicated in the causation of CAUTI. The most frequently isolated bacterial CA-UTI pathogens in the present study were *E. coli* 25 (9.3%) followed by *Enterococcus species, 20 (7.4%) Klebsiella species 19* (7%) which is lower in prevalence compared to studies conducted in India *E.coli* (34.85%), *Klebsiella* (19.7%) (25); Egypt *E. Coli* (35%) (41), India *E.coli* (38.71-57%) *Klebsiella* (17.74-20%) (29, 39).The variation in diversity of bacterial pathogen might be because of differences in environmental and climatic conditions, catheter duration and the colonising organisms' uniqueness to hospital environments (25).

6.2 Prevalence of *Enterococcus* species

The current study revealed that 7.4% of *Enterococcus* species were isolated from the study participants with urinary catheter. This finding is comparable with previous studies conducted in Serbia 9% (22), India 6. %, 6.06% and 6.7% (25, 29, 40) and in Ethiopia 5.5% (2). Compared to the current finding, lower *Enterococcus* species were reported in India (30, 39) 1.5% and 3.23% Egypt (42) 0.74%.

On the other hand, higher *Enterococcus* prevalence was documented in studies conducted in USA 12%-18% (35 38), Egypt12.8%-16.4% (41, 44), and Hungary 14.9% (37). Differences in the prevalence of *Enterococci* isolates might be due to geographic variations and methodological variations in bacterial identification (52).

Multivariate analysis showed that factors such as intermittent insertion of catheter, marital status, history of previous UTI and type of admission wards were independent predisposing risk factors for CAUTIs. Current finding is in line with study done in Indonesia (41) that Old aged > 50 years and duration of catheterisation had strong association with increased risk of developing CA-UTI.

6.3 Antibiotic Susceptibility of *Enterococcus* Isolates

In the current study, the prevalence of vancomycin resistant *Enterococcus* in patients with indwelling catheter is 20% using E-test method. The finding is in line with studies conducted in Jimma, Ethiopia 22.7% (21), Arbaminch, Ethiopia 22% (34) Saudi Arabia 17.3%, (19) and India 16.19% (28) but higher compared to studies done in India5 .7%, (27), USA 6.3%, (38) Hungary Budapest 6,1% (37**)** India 7 %, (20) and in Ethiopia, Black Lion 6.7%. (16). The current finding is lower than studies conducted in Egypt 85.7%), (41) and India 36.1% (20). Majority of the previous studies the sensitivity test was performed by using disc diffusion test.(21,34,16,20) but some are done using molecular AST tests (37,38)) the rest were using E-Test (28,41,27)

6.4 Knowledge of health professionals on CA-UTI

The current study assessed the knowledge and awareness of health professionals on catheter associated urinary tract infection. Accordingly, 94.2% of the participants were aware that CAUTI is one of the hospital acquired infection. This is comparable with a study done in India 82.1% (45). Knowledge on catheter associated drug resistance is 80.2% in the current finding which is higher compared to study conducted in Nigeria 49.2% (46). In the present study, 72.7% of participants argued that following strict aseptic precautions during urinary catheterization helps to prevent CA-UTI. Similarly, a study in India reported 95.8% (45). Also 64.5% of participants' aware that hand washing before and after handling the catheter site with antiseptics is mandatory for patient's safety. But lower with studies done in India 96.8% (45). More than half (57%) of the participants confirmed that regular educational training improves knowledge of health professionals on CA-UTI. The current finding is lower than study performed in India 94.7% (45).

Multivariate analysis in logistic regression showed that there was no significant association of age, gender, profession, education, and years of experience in health care professional with the level of knowledge among the study participants. Similar study with the current finding is comparable concerning demographic data do not have statistically significant associations (32). The variation of knowledge level among health professions in the study compared to others might be due to various reasons including study protocols, sample size, profession diversity, methodology and duration of study (45).

7. STRENGTHS AND LIMITATIONS

7.1 Strengths

1. Vancomycin resistant *Enterococcus* prevalence was performed by E-test method to increase the accuracy and precision of the prevalence rate
2. The research is the first in Addis Ababa city used as a base line data
3. Urine sample was collected from admitted and catheterized clients since these groups are highly vulnerable for hospital procedural acquired infection.

7.2 Limitations

1. *Enterococcal* biofilm production was not performed due to lack of reagents (culture media).
2. Sample size was too small in order to generalize the results to the whole population.
3. Unable to detect of *Enterococci* in species level as well as antimicrobial resistance profile against each isolated species level.
4. Reagents and some antibiotic were not neither available in the country nor imported abroad easily.
5. Urine sample was not collected before administration of antibiotics. Antibiotics may hinder the growth of bacteria during culturing.

8. CONCLUSIONS AND RECOMMENDATIONS

8.1 Conclusion

The overall prevalence of catheter-associated urinary tract infection by bacterial pathogen was 31.9%. More than 2/3 of the isolates were gram negative bacteria (69.8%) whereas gram positive bacteria constituted (30.2%). Most predominant etiologies of CAUTIs among catheterized patients were *Escherichia coli* 25 (9.3%), *Enterococcus* species 20 (7.4%) and *Klebsiella* species 19(7%).

Drug resistance observed in penicillin (95%), Doxycycline (55%), erythromycin (50 %); Vancomycin disc diffusion (45%) ceftriaxone (40%) which showed greater than 40% of resistance rate. In contrast, meropenem (20%) and ciprofloxacin (25%) shows less resistance rate below 30% resistance. Susceptibility test for E-Test and disc diffusion test was 70% and 40% respectively.The study revealed that majority of health professionals (94.2%) had knowledge on CAUTI and were aware of the existence of multi drug resistance, whereas most of the health professionals were not aware of bacterial biofilm formation which will lead to VRE and emergence of other drug resistance. Periodic monitoring of patients safety and screening of antimicrobial resistance patterns improves the overall clinical outcome of the patients

8.2 Recommendations

1. Long term and intermittent catheterization is the risk factor for CAUTI hence shortening of duration will prevent the prevalence. Health professionals need to consider risk minimization protocols
2. Routine practice of drug resistance screening need to be practiced in hospital and regional level.
3. Continuous training on infection prevention might increase level of knowledge and awareness of health professionals on CA-UTI and spread of drug resistance.

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ANNEXES

9.1 Annex-I Research project Information

Title of the project- Catheter associated vancomycin resistant *Enterococcus* at Yekatit 12 hospital medical college admitted patients, Addis Ababa, Ethiopia.

Name of the principal investigator: Ayelign Derebe (BSc, MSc)

Name of the organization: Addis Ababa University, college of health science, Department of Microbiology, Parasitology and Immunology.

Introduction:

This information is prepared by principal investigator to determine the prevalence of Catheter associated urinary tract infection and vancomycin resistant pattern of *Enterococcus species* among admitted patients with indwelling urinary catheter as well as to assess the level of knowledge and awareness of health care professionals about catheter associated UTI and drug resistances in Yekatit 12 Hospital medical college Addis Ababa, Ethiopia, 2020.

Purpose:

To determine the prevalence of catheter associated vancomycin resistant *Enterococcus* species circulating and spreading in hospital admission wards and to assess the knowledge and awareness of health care workers about device associated UTI, drug resistance and hospital acquired infections.

Procedure:

The study participants are kindly invited to take part in the research project if they are willing to participate in the study, they need to understand and sign the agreement form. For laboratory examinations, they were requested to provide urine sample from catheter or catheter tips and the specimens were collected following standard protocol to identify the etiology of catheter associated urinary tract infections. The laboratory examination results were kept confidential using coding system whereby no one will have access to the result. For isolated pathogens, physicians attending the clients were communicated for further intervention and selecting the drug of choice for the infection. The health care workers were requested to participate and to fill the lists of the data properly on the questionnaires information sheet provided.

Risks and discomforts

There is no major known risk. The risk related to the data management and confidentiality was managed as per the ethical consideration and based on Helsinki declaration.

Benefits

There may not be a direct benefit that you will gain from participating in this study. However, you will be able to know microorganisms responsible for your illness and the best antibiotic treatment of choice for your illness. Besides, Health facilities, MOH, and research laboratories will be benefit from the findings. The potential benefit to society is significant if insight into the susceptibility pattern of the key device associated pathogens against treatment of choice. Moreover it will be used for indicator of the current status of this bacteria and also start up for surveillance purposes.

Incentives:

You will not be provided any incentives to take part in this research. Only the sample collector was will be paid compensation per sample.

Confidentiality-

Any information obtained during this study was kept confidential and assured by avoiding use of any identifier and information recorded with code number.

Voluntary participation:

Participation on this study was voluntary and have the right to refuse at any time. Your decision will not result in any penalty or loss of benefits to which you are entitled. Your decision will not put at risk any present or future services to which you are entitled. You may ask questions now and in the future if you do not understand something that is being done.

Here is an address of principal investigator, who you may contact,

Mr. Ayelign Derebe, +251911881040, ayelign1977@gmail.com

9.2 Annex II Consent Form

Participant Code Number_____

I informed and understood the aim of the above mentioned research project and am willing to participate in the study of "**Catheter associated vancomycin resistant *Enterococcus* at Yekatit 12 hospital medical college admitted patients, Addis Ababa, Ethiopia**". I have been informed that there is no risk by participating on the project. In addition I have been told all the information collected throughout the research process will be kept confidential. Moreover I have been well informed of my right to keep hold of information, decline to cooperate and make withdrawal from the study.It is therefore with full understanding of the situation that I gave the informed consent voluntarily to participate in this research. In addition, I have had the opportunity to ask questions about it and received clarification to my satisfaction. I have also been informed that the project have many benefit in the country level.

Study participant name_____Signature_____Date_____

Data collector's name _____Signature_____

Eyewitness name----------signature----------

Date of data collection_____Time started_____Time finished_____

Thank you in advance for your participation in this study!!

Annex-II Consent form (Amharic Version)

የስምምነት ቅጽ

መለያ ቁጥር……

የጥናት ተሳታፊ ስምምነት ማረጋገጫ ፊርማ……

ይህንን የተሳታፊነት መረጃ እና ስምምነት ቅጽ እኔአንብቤዉ (ወይም አንድ ሰው ለእኔ አንብቦልኝ) እንደተረዳሁት በአንድ ምርምር ርጥናት ላይ ለመሳተፍ እየተጠየኩኝ ነዉ:: እኔም ጥያቄዎችን የመጠየቅ አጋጣሚ ነበረኝ የተሰጠኝ ምላሽ እና ማብራሪያ በቂ ነዉ:: እኔ በፈቃደኝነት በዚህ ጥናት ውስጥ ለመሳተፍ ተስማምቻለሁ:: እኔ ይህን ቅጽ በመፈረሜ ማንኛውም ሕጋዊ መብቴን አሳልፌ እየሰጠሁ አይደለም:: የዚህ የተሳታፊነት መረጃ እና ስምምነት ቅጽ ቅጂ ይሰጠኛል::

<u>የተመራማሪው / የምርምርሠራተኛ.</u>

እኔ ተመራማሪው ለምርምሩ ተሳታፊዎች ስለምርምሩ በቂ እና ግልጽ ማብራሪያ ከሰጠሁ በኃላ መስማማታቸዉን በፊርማ እንዲያረጋግጡልኝ ጠይቄአለሁ::

የጥናት ተሣታፊዉ ሙሉ ስም_____

ፊርማ: _____ **ቀን:** _____

የተመራማሪዉሙሉስም:_____

ፊርማ:_____**ቀን**_____

በጥናትና ምርምሩ በመሳተፍዎ በጣም እናመሰግንዎታለን!!

9.3 Annex-III Information survey questionnairesheets (1)

The following detail information listed below are required to determine the prevalence of catheter associated vancomycin resistant *Enterococcus* among hospitalised clients and to assess the general knowledge and awareness of health care workerson device associated antimicrobial

resistance in hospital settings at Yekatit 12 hospital medical college.Therefore, we kindly request your honest response to this survey questionnaire. Indeed, your participation is voluntary. The survey may take about 10 minutes to fill out. Your responses will be completely anonymous.

SECTION 1: BACKGROUND INFORMATION			
S.NO	**ITEMS**	**RESPONSE**	**Remark**
A	Client Unique code_____		
B	Client Medical registration No_____		
101	How old are you?	16 -24Year 25-45year 46-65 year Above 65 year	
102	Gender?	1. Male 2. Female	
103	What is Your Marital status?	1. married 2. Single 3. Divorced	
104	What is Your Educational status?	1. Illiterate 2. 10/12 complete or under 3. Certificate/diploma 4. degree and above	

SECTION 2: CLIENTS CLINICAL STATUS INFORMATION

201	What is the reason for your admission in this ward?	1. surgery case 2. delivery 3. Medical care 4. emergency care	
202	Do you have any previous chronic disease before? If yes what was it?	1. diabetes 2. hypertension 3. HIV, Cancer&STI	
203	Do you have a history of previous UTI infection?	1. yes 2. No	
204	Do you have a history of previous urinary catheterization?	1. Yes 2. No	
205	How many days did you spend with inserted urinary catheter?	1. <3 days 2. 4—9 days 3. 10 -30days 4. Intermittent insertion	
206	Do you have a any previous procedures around your genital area	1. Yes 2. No	
207	Do you have a fever during your stay in the hospital?	1. Yes 2. No	
208	Do you have a urinary urgency or frequency during your stay in hospital?	1. Yes 2. No	
209	Do you have back pain around your back and pelvic area?	1. Yes 2. No	
SECTION 3: CLIENTS CURRENT TREATMENT INFORMATION			
301	Are you on antibiotics Currently?	1. Yes 2. No	
302	Duration of antibiotic administration	1. 3 days 2. 7 days 3. 14 days 4. >14 days	
304	Any other medication for chronic illness	1. Yes 2. No	

9.4 Annex-IV Informationsurvey questionnairesheet (2)

The following detail information listed below are required to assess the general knowledge and awareness of health care professionals on catheter associated urinary tract infection and

antimicrobial resistance in hospital settings at Yekatit 12 hospital medical college. Therefore, we kindly request your honest response to this survey questionnaire. Indeed, your participation is voluntary. The survey may take about 10 minutes to fill out. Your responses will be completely anonymous.

Part I-socio-demography information			
S/N	**questions**	**Categorical Responses**	
Q 401	Which category of age groups are you?	1. <24Year 2. 25-40 year 3. 41-55 year	
Q 402	gender	1. Male 2. Female	
Q 403	What is Your Marital status	1. married 2. Single 3. Divorce	
Q 404	What is your current Educational status	1. Nurse 2. Health officer 3. Doctor	
Q 405	How long is your Work experience?	1. < 1 year 2. 1—5 years 3. 6—15 years 4. >16 years	
Part-II Knowledge assessment			
Q 406	How deep are you aware about health care associated infection	1. Yes 2. No	
Q 407	Do you have any awareness on catheter associated urinary tract infection in hospital?	1. Yes 2. No	
Q 408	did you have understanding on antimicrobial drug resistance?	1. Yes 2. No	
Q 409	Do you have a knowledge aboutVancomycin Resistant entrococcus?	1. Yes 2. No	
Q 410	Do you know about bacterial Biofilm formation?	1. Yes 2. No	

Part-III assessment to practice			
Q 411	How strong is your adherence to aseptic procedures and patient's safety practice?	1. Very strong 2. Strong 3. Less strong 4. Not measured	
Q 412	How much is the Supply of consumable safety materials for hospital environments?	1. Available always 2. Limited availability 3. Not available	
Q 413	Are universal patient safety guidelines available?	1. Yes 2. No	
Q 415	Do you have trainings on infection prevention and patient's safety?	3. Yes 4. No	
Q 416	Do you have feedbacks from clients on safety issue?	1. Yes 2. No	
Q 417	Do you wash your hands before and after catheter insertion?	1. Yes 2. No	

THANK YOU SO MUCH FOR YOUR COOPERATION!!.

Annex-V Guardians consent

I hereby confirm that the investigator has informed me about the nature of the study. I have also received, read and understood the above written information regarding the study. I am aware

that the results of the study, including personal details regarding my age, sex, educational status etc.… will be anonymously processed into a research report. I may, at any stage, withdraw my consent and participation in the study. I had sufficient opportunity to ask questions and of my own free will declare myself prepared to participate in the study.

Statement of Parent or Guardian

My relative appears to understand the research to the best of his or her ability and has agreed to participate.

Guardian's

Name_____Signature_____Date_____

Participant's Name _____signature_____Date _____

Name of PI _____signature _____Date _____

I, **Ayelign Derebe Kindie** (investigator) herewith confirm that the above participant has been informed fully about the nature of the above study.

Witness's name _____

Witness's signature _____

Date _____

Annex-V

<u>የወላጅ/የሞግዚት</u> የስምምነት ማረጋገጫ ቅጽ

መለያ ቁጥር _____

የጥናት ተሳታፊ ስምምነት ማረጋገጫ ፊርማ _____

ይህንን የተሳታፊነት መረጃ እና ስምምነት ቅጽ እኔ አንብቤዉ (ወይም አንድ ሰው ለእኔ አንብቦልኝ) እንደተረዳሁት በአንድ ምርምር ጥናት ላይ ለመሳተፍ እየተጠየኩኝ ነዉ:: እኔም ጥያቄዎችን የመጠየቅ አጋጣሚ ነበረኝ የተሰጠኝ ምላሽ እና ማብራሪያበ ቂነዉ:: እኔ በፈቃደኝነት በዚህ ጥናት ውስጥ ለመሳተፍ ተስማምቻለሁ:: እኔ ይህን ቅጽ በመፈረሜ ማንኛውንም ሕጋዊ መብቴን አሳልፌ እየሰጠሁ አይደለም:: የዚህ የተሳታፊነት መረጃ እና ስምምነት ቅጽ ቅጂ ይሰጠኛል::

<u>የወላጅ ወይም የሞግዚት መግለጫ</u>

ይህ የጥናት ምርምር ለዘመዴ ጠቃሚ እንደሚሆን በመረዳት በጥናቱ እንዲሳተፍ መስማማቴን በፊርማዬ አረጋግጣለሁ፡፡

<u>ተመራማሪው / የምርምር ሠራተኛው</u>

እኔ የምርምር ሠራተኛው ለምርምሩ ተሳታፊዎች ስለምርምሩ በቂ እና ግልጽ ማብራሪያ ከሰጠሁ በኃላ መስማማታቸዉን በፊርማ እንዲያረጋግጡልኝ ጠይቄአለሁ::

<u>የጥናት ተሣታፊዉ ወላጅ/ ሞግዚት</u>

ሙሉ ስም:_____

ፊርማ: _____

ቀን: _____

<u>የተመራማሪዉ</u>

ሙሉ ስም:_____

ፊርማ:_____

ቀን_____

በጥናትና ምርምሩ በመሳተፍዎ በጣም እናመሰግንዎታለን!!

Annex-VIAST result interpretation in Kirby-Bauer disc diffusion methods and E-Test methods

AST result interpretation (Kirby-Bauer disc diffusion method)

Antimicrobial agents	Disc potency	Zone of inhibition			Remarks and notices
Vancomycin	30 µg	≤ 14	15-16	≥ 17	For the reading indicated as I

	R	**I**	**S**	and R, E-Test will be used for double check confirmation

AST result interpretation (E-Test method)

Antimicrobial agents	Disc potency	Reading MIC			Remarks
Vancomycin E-test	.016-256 µg	**S**	**I**	**R**	Result interpretation
		≤4	**8-16**	32 ≥	Reading 6 will round to 8, reading 18,20,24,26,28,30 will round to 32

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: AyelignDerebeKindie

Signature: _____ Date of submission: _____

This thesis has been submitted with our approval as advisors.

Advisor: Signature: _____**Date**: _____

Co-Advisor: Signature: _____**Date**: _____

Place: Addis Ababa, Ethiopia.