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Fecal Carriage of Extended-Spectrum β-Lactamase and Carbapenemase-Producing *Enterobacterales* among Oncology Patients at St. Paul's Hospital Millennium Medical College, Addis Ababa, Ethiopia

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This is to certify that the thesis prepared by Tsedale Woldu entitled:Fecal Carriage of Extended-Spectrum β-Lactamase- and Carbapenemase-Producing *Enterobacterales* among oncology patients at St. Paul's Hospital Millennium Medical College, Addis Ababa, Ethiopia,and submitted in partial fulfillment of the requirements for the Master of Science degree in Clinical Laboratory Sciences (Diagnostic and Public Health Microbiology specialty track) complies with the regulations of the University and meets the accepted standards with respect to originality and quality.

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Abbreviations

5FU	5-Fluorouracil
ACT	Adriamycin and Cyclophosphamide Therapy
AMR	Antimicrobial Resistance
ATCC	American Type Culture Collection
CAPOX	Capecitabine and Oxaliplatin
CLSI	Clinical and Laboratory Standards Institute
CPE	Carbapenemase Producing *Enterobacterales*
DDST	Double Disk Synergy Test
ECDC	European Centre for Disease Prevention and Control
ESBL	Extended-Spectrum β-Lactamase
FOLFOX	5-Fluorouracil, Leucovorin, and Oxaliplatin
GI	Gastrointestinal
MHA	Mueller Hinton Agar
MDR	Multidrug-Resistant
MIL	Motility Indole Lysine
PCR	Polymerase Chain Reaction
SPHMMC	St. Paul's Hospital Millennium Medical College
TSI	Triple Sugar Iron
WAAR	World Alliance Against Antibiotic Resistance
WHO	World Health Organization

Abstract

Introduction: Antimicrobial resistance (AMR) is a major public health problem worldwide, with extended-spectrum β-lactamase (ESBL) and Carbapenemase-producing *Enterobacterales* contributing significantly to multidrug resistance. These organisms colonize the gastrointestinal tract and act as a reservoir for serious infections. Oncology patients are especially at risk because of immunosuppression and repeated exposure to antibiotics.

Objective: To determine the fecal carriage rate of Extended-spectrum β-lactamase and carbapenemase-producing *Enterobacterales* among oncology patients at St. Paul's Millennium Medical College Hospital, Addis Ababa, Ethiopia

Method:A cross-sectional study was conducted at the oncologic unit of St. Paul's Hospital Millennium Medical Collegefrom January to March 2025. Oncology patients were enrolled using a consecutive sampling technique. Stool samples were screened using chromogenic media for ESBL- and Carbapenemase-producing *Enterobacterales*. Isolates were identified using conventional biochemical tests. Kirby–Bauer disk diffusion method was used to determine antimicrobial susceptibility of isolates. Production of ESBL enzyme was confirmed byusing the combination disk method, while production of carbapenemase was confirmed using the modified carbapenem inactivation method (mCIM). Data were entered using Kobo Toolbox and analyzed using SPSS version 27. Univariate and bivariate analyses were performed, and associations were assessed using theap-value of less than 0.05 was regarded as statistically significant.

Result: Among 226 cancer patients, the intestinal carriage rate of ESBL-producing *Enterobacterales*(ESBL-PE)was 67.3% (152/226), *and* carbapenemase-producing *Enterobacterales*(CPE)was 17.7% (40/226), predominantly *E.coli*. ESBL-Producing isolates showed the highest resistance to ciprofloxacin (52.5%) and cefepime (43%), while amikacin had the lowest resistance (4.4%). CPE isolates exhibited very high resistance to ceftazidime and cefotaxime (97.7%), cefepime (95.5%), and ciprofloxacin (93.3%), with comparatively lower resistance to amikacin (55.5%). 14.6% ESBL-PE and 57.8% CPE isolates were multidrug-resistant. Repeated antibiotic treatment and indwelling devices were associated with ESBL-PE carriage, while age, male sex, and comorbidities were associated with CPE carriage ($p<0.05$).

Conclusion: Most isolates recovered from fecal specimens of oncology patients were resistant to multiple antibiotic classes, and some showed reduced susceptibility to carbapenems. These findings underscore the need for routine screening, strict infection prevention, and judicious antibiotic use to protect vulnerable patients and curb the spread of antimicrobial resistance.

Keywords: *Enterobacterales*, Fecal carriage, ESBL, CPE, Oncology

1. Introduction

1.1 Background

Antibiotic resistance is currently a global public health concern that affects both human and animal health. The rapid development and global spread of multidrug-resistant microorganisms that cause common diseases and resist treatment are particularly concerning. According to the World Health Organization (WHO), Antimicrobial Resistance is among the top ten worldwide public health risks that humanity faces(1–3).

Enterobacterales are the primary bacterial species responsible for producing Extended-Spectrum β-lactamases (ESBLs)(4). The gastrointestinal tract of humans frequently contains*Enterobacterales.* These organisms can cause a range of extra-intestinal infections in the lower and upper urinary tracts, bloodstream, central nervous system, pelvis or abdomen, and wounds. *Escherichia coli*and *Klebsiella pneumoniae* are the most significant *Enterobacterales*linked to production of ESBL enzymes(4).

ESBL-producing *Enterobacterales* (ESBL-PE) frequently show resistance to numerous antibiotics, and co-resistance to others that are used for treatment, including aminoglycosides, fluoroquinolones, and trimethoprim. As a result, options of treatment for critically sick patients are limited, leading to increased morbidity and mortality rates. Therefore, these organisms are creating a significant public health problemglobally. The prevalence and distribution of ESBLs vary widely across geographic regions, both between countries and within countries between hospital and community settings(4,5).

According to the World Union against Anti-Microbial Resistance (WAAR), antimicrobials may eventually lose their effectiveness. This could be the result of both improper use of antimicrobial medications and self-medication. The use of these restorative operators is driven by the rise of multidrug-resistant bacterial species, and in reality, a few of them are resistant to all available antimicrobials(6).

The growing prevalence of carbapenem-hydrolyzing enzymes, such as KPC-2 (*Klebsiellapneumoniae* carbapenemase-2), and β-lactamase enzymes, such as NDM-1 (New

Delhi metallo-β-lactamase-1), has become a major global public health concern. This is largely due to the limited therapeutic options available for treating infections caused by microorganisms harbouring these resistance mechanisms (7). The 2019 report by the CDC on antimicrobial resistance threats categorized ESBL-producing *Enterobacterales* (ESBL-PE) and carbapenemase-producing *Enterobacterales* (CPE) among the most critical antimicrobial-resistant pathogens responsible for severe infections in humans(8). The WHO also designated CPE and ESBL-PE as critical-priority antibiotic-resistant pathogens, underscoring the urgent need for enhanced surveillance, research efforts, and the development of effective therapeutic options against these organisms.According to WHO estimates, antimicrobial-resistant bacteria cause 700,000 fatalities worldwide each year. A review on antimicrobial resistance also predicts that deaths related to superbugs could reach 10 million annually by 2050 if treatment strategies are not changed(9).

1.2 Statement of the problem

Multidrug resistance among Gram-negative bacteria is increasing globally, affecting both community and hospital-acquired infections (9). Within the *Enterobacterales* family, resistance to extended-spectrum β-lactams and carbapenems has become a major clinical concern. Extended-spectrum β-lactamase-producing *Enterobacterales* (ESBL-PE) and carbapenemase-producing *Enterobacterales* (CPE) are critical priority pathogens that significantly limit treatment options and worsen patient outcomes (10).

The burden of cancer further intensifies this problem. In 2018, there were an estimated 9.5 million cancer-related deaths and 18 million new cases worldwide(11). In Ethiopia, cancer contributes to approximately 5.8% of all deaths, with an estimated 60,960 new cases and more than 44,000 cancer-related deaths occurring each year (12). Cancer patients, particularly those with haematological malignancies, are highly susceptible to infections caused by multidrug-resistant Gram-negative bacteria such as *Acinetobacter baumannii*, *Pseudomonas aeruginosa*,*Escherichia coli*, *Klebsiella pneumoniae*, and *Stenotrophomonas maltophilia* due to immunosuppression, mucosal damage, and invasive procedures (11).

Broad-spectrum β-lactam antibiotics are commonly used for empirical treatment in these patients; however, the increasing prevalence of ESBL-PE and CPE, often resistant to multiple drug classes, poses a serious therapeutic challenge (13,14). The spread of ESBL-producing organisms is largely driven by plasmid-mediated resistance, facilitating rapid transmission across bacterial species (15).

Gastrointestinal colonization with ESBL-PE plays a key role in subsequent infections and transmission. Colonized patients may develop invasive infections, particularly when immunocompromised, and can serve as reservoirs for the spread of resistance (16). This risk is heightened in cancer patients due to chemotherapy-induced immune suppression and disruption of intestinal barriers, promoting bacterial translocation (17). Colonization has been strongly associated with increased risk of infection, with additional risk factors including advanced age, inappropriate antibiotic use, and poor hygiene practices (18–20).

Despite the growing threat, data on fecal carriage and antimicrobial resistance patterns of ESBL-PE and CPE among cancer patients are limited in Ethiopia, particularly in Addis Ababa. At St. Paul's Hospital Millennium Medical College (SPHMMC), oncology patients frequently present with febrile illnesses that may be linked to underlying colonization by resistant organisms.

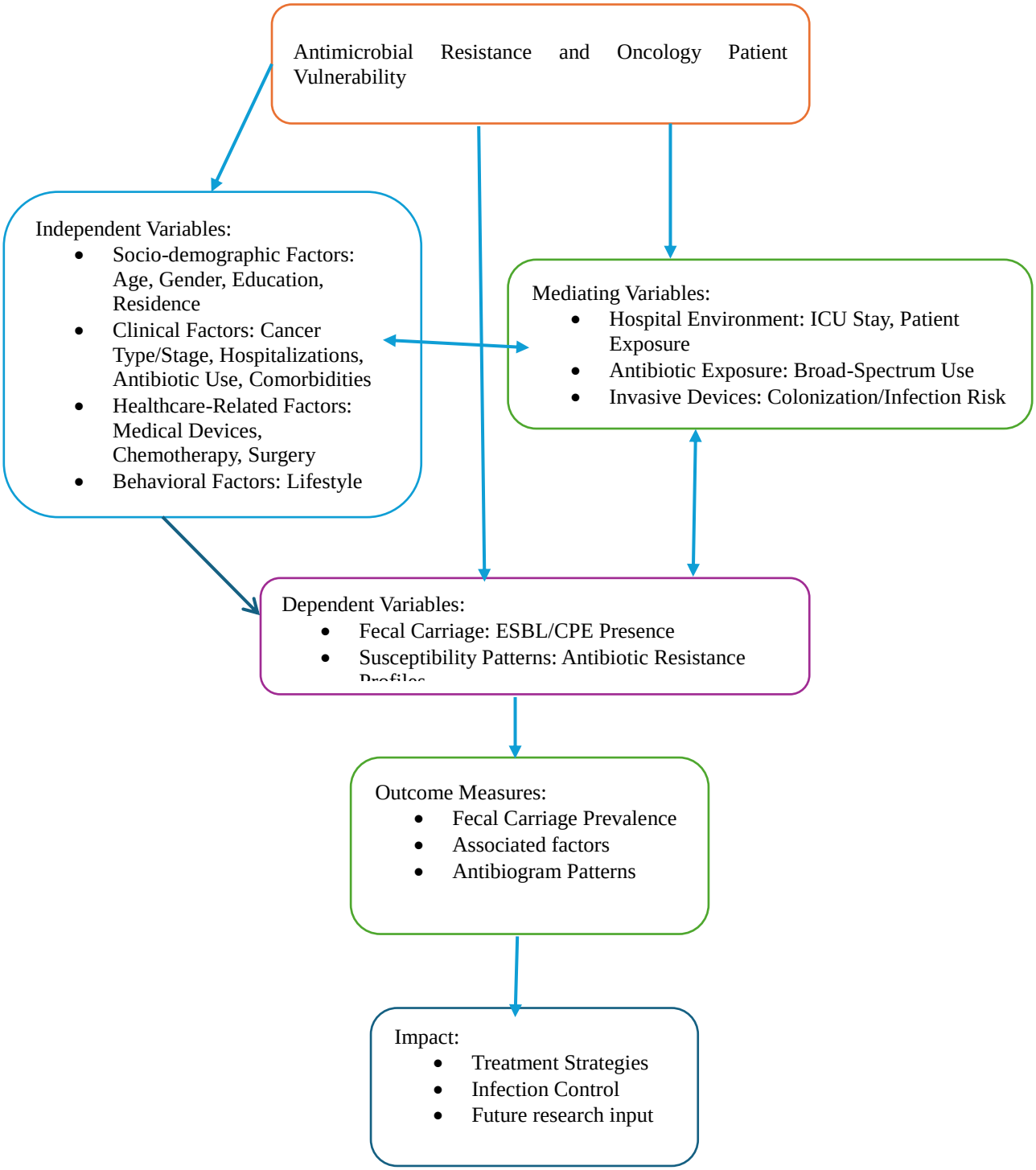
Therefore, this study aims to determine the prevalence of fecal carriage of ESBL- and carbapenemase-producing *Enterobacterales* and to assess their antimicrobial susceptibility patterns among oncology patients at SPHMMC, Addis Ababa, Ethiopia.

1.3 Significance of the study

The researchimpact lies in addressing the growing concern of multidrug-resistant *Enterobacterales*, such as ESBL-PE and CPE, which pose serious health risks due to limited treatment options. By assessing the prevalence, associated factors, &antibioticresistancepatterns of these resistant strains betweenoncology patients at SPHMMC, the study aims to inform more effective empirical treatment strategies, reduce morbidity and mortality, and prevent the spread of these pathogens. The findings will improve patient access to targeted treatments, enhancing outcomes and care quality. Healthcare workers gain evidence-based guidance for decisions and infection control, reducing exposure to resistant strains. Policymakers receive data to shape

guidelines, strengthen stewardship, and implement interventions at hospital and community levels, supporting a coordinated response to antimicrobial resistance.

Conceptual framework



2. Literature review

2.1*Enterobacterales*

Enterobacterales comprise a diverse group of Gram-negative, rod-shaped bacteria that are commonly found in the gastrointestinal tract of mammals but can also survive and proliferate in environmental reservoirs such as soil, water, and plants.Theyare facultative anaerobic, non-spore-forming Gram-negative bacilli.This diverse group includes genera such as *Escherichia*, *Citrobacter*, *Enterobacter*, *Proteus*, *Hafnia*, *Klebsiella*, *Providencia*, and *Serratia*. It also encompasses several important enteric pathogens, including *Salmonella* spp., *Shigella* spp., *Yersinia enterocolitica*, and pathogenic strains of *Escherichia coli*(21). Members of the order *Enterobacterales* are generally oxidase-negative, indicating the absence of cytochrome c oxidase activity. They characteristically ferment glucose with the production of acid and are capable of reducing nitrates to nitrites, features that are commonly used for their laboratory identification(22).

Enterobacterales are associated with two broad categories of infections: intestinal and extraintestinal diseases. Intestinal infections are primarily transmitted through the fecal–oral route, either via direct person-to-person spread or, more commonly, through the ingestion of contaminated food and water containing pathogenic organisms.Endogenous infection can result from the translocation of bacteria from the intestinal tract into sterile body sites, particularly among immunocompromised individuals such as cancer patients, those with liver cirrhosis, and patients undergoing chemotherapy. In this population, antibiotic-resistant Gram-negative bacteria, especially carbapenem-resistant and broad-spectrum β-lactam-resistant strains of *E. coli*, *K. pneumoniae*, *A. baumannii*, *P. aeruginosa*, &*S. maltophilia*, are among the most significant causes of infection & are associated with considerable morbidity, mortality, and therapeutic difficulties (22).

2.2 Oncology Patients as a High-Risk Group

Oncology patients are particularly vulnerable to infections due to several factors, including immunosuppression, the effects of chemotherapy, and prolonged hospital stays. Chemotherapy targets rapidly dividing cancer cells but also adversely affects hematopoietic cells, leading to neutropenia, which significantly compromises the immune response. This immunocompromised state increases susceptibility to infections, particularly from resistant pathogens such as CPE.

Studies have shown that patients undergoing intensive chemotherapy are at a higher risk for developing infections, with a notable prevalence of CPE in this population, often exacerbated by previous antibiotic exposure and prolonged hospitalization(23). Furthermore, the use of central venous catheters and other invasive procedures increases the risk of healthcare-associated infections, making timely diagnosis and treatment critical(24).

The impact of infections caused by resistant *Enterobacterales* on cancer patients is profound, leading to increased morbidity and mortality. Infections with CPE have been associated with mortality rates as high as 60% among neutropenic patients with hematological malignancies(23). These infections complicate treatment regimens and can lead to delays in necessary cancer therapies, further jeopardizing patient outcomes. The burden of antibiotic resistance not only complicates clinical management but also poses significant challenges in resource-limited settings, where treatment options may be severely restricted(25). Consequently, the emergence of multidrug-resistant organisms (MDROs) in this vulnerable population highlightsthe critical need for efficient antimicrobial stewardship and infection control strategies to reduce these dangers (26).

2.3 Mechanisms of action and resistance of antibacterial drugs

Antimicrobial resistance (AMR) refers to the ability of bacteria to resist the action of antimicrobial agents, thereby reducing the effectiveness of treatments and complicating infection management that were previously effective against them when used at standard therapeutic doses(27). Antibacterial drugs act by disrupting key microbial processes, including nucleotide synthesis (thereby inhibiting DNA and RNA formation), protein synthesis, and cell membrane integrity, or by interfering with enzymatic pathways involved in bacterial cell wall synthesis(28). Bacteria can acquire multiple mechanisms of resistance, including (i) reduced uptake of antibiotics, (ii) production of hydrolytic enzymes such as β-lactamases that inactivate antimicrobial agents, (iii) alteration of drug target receptors, (iv) active efflux pumps of antibiotics via membrane-associated efflux pumps that reduce intracellular drug concentrations, (v) modification of enzymatic pathways leading to decreased bacterial susceptibility, and (vi) loss of intracellular enzymes required for activation of prodrugs. Among these mechanisms, the production of β-lactamases is the most frequently encountered(28).

2.3.1 Mechanism of action in β-lactam resistance

The widespread use of β-lactam antibiotics has been strongly linked to the emergence of bacterial resistance mediated by β-lactamase enzymes(29). β-lactamases are bacterial enzymes that confer resistance by hydrolyzing the β-lactam ring of these antibiotics, thereby irreversibly inactivating the drug through cleavage of the amide bond (30). Currently,two classification schemes areused for β-lactamases: functional (Bush-Jacoby classification) and molecular (Amblerclassification)(31,32).The functional classification system categorizes β-lactamases according to their ability to hydrolyze different classes of β-lactam antibiotics and their susceptibility to β-lactamase inhibitors(32).In contrast, the widely adopted molecular classification proposed by Ambler is based on amino acid sequence homology and classifiesβ-lactamases into four classes. Classes A, C, & D are serine β-lactamases that utilize a serine residue at the active site for hydrolysis of substrates, whereas class B enzymes, known as metallo-β-lactamases, depend on divalent zinc ions to mediate β-lactam hydrolysis (31).Production of beta-lactamases can be inducible or constitutive (33). In Gram-positive bacteria, enzyme production is typically inducible, with high levels of β-lactamase being synthesized in response to exposure to antibiotics (34). In contrast, Gram-negative bacteria often exhibit constitutive production, whereby the enzyme is expressed continuously even in the absence of antibiotic pressure (35). Nevertheless, AmpC β-lactamases in certain Gram-negative organisms are inducible, particularly in species such as *Serratia*, *Enterobacter*, *Escherichia*, and *Citrobacter* (36).

2.3.2 Mechanism of action of Carbapenemase resistance.

Carbapenemase production represents one of the primary mechanisms of resistance in carbapenemase-producing *Enterobacterales* (CPE). In addition, these organisms may concurrently produce extended-spectrum β-lactamases (ESBLs) and/or AmpC enzymes, and may also exhibit reduced permeability due to loss or alteration of outer membrane porin proteins (OMPs)(37). Carbapenemases are β-lactamase enzymes capable of hydrolyzing carbapenems and are classified into Ambler classes A, B, and D. Within *Enterobacterales*, KPC is a well-characterized class A carbapenemase. The KPC enzyme hydrolyzes a broad range of β-lactam antibiotics, including penicillins, cephalosporins, and carbapenems; however, its activity can be restrained by inhibitors ofβ–lactamase like tazobactam & clavulanic acid, while it remains

resistant to other antimicrobial classes(38). In *Enterobacterales*, carbapenem resistance is believed to arise from the loss or decreased expression of outer membrane porin (OMP) proteins, either alone or in combination with the production of β-lactamases such as ESBLs and AmpC enzymes. In *Klebsiella pneumoniae*, the principal porins include OmpA and OmpK. Carbapenemase-producing "superbugs," particularly those producing NDM-1 and its variants, represent a major global public health concern. NDM-1 can hydrolyze a broad range ofβ-lactam antibiotics, including carbapenems, which are often reserved as last-line agents for treating multidrug-resistant bacterial infections (39).

2.4 Epidemiology of β-lactamase- and Carbapenemase-producing broad-spectrum

A hospital-based study in India reported that 39 out of 60 intensive care unit patients (65%) had ESBL-producing *Enterobacterales* (ESBL-PE) detected in their stool samples. Among these isolates, 22 (56%) were identified as *Escherichia coli* and 17 (43%) as *Klebsiella* species(40).

A Korean study on hospitalized patients showed that 37 out of 97 individuals (38.1%) were colonized with ESBL-PE based on rectal swab analysis. Among these, *Escherichia coli* was identified in 17 patients (17.5%), while *Klebsiella pneumoniae* was detected in 14 patients (14.4%) and *Klebsiella oxytoca* in 2 patients (2.1%). In addition, co-colonization with both *E. coli* and *Klebsiella pneumoniae* was observed in 4 patients (4.1%)(41). Another study from Tanzania also reported comparable findings, with an overall ESBL colonization rate of 34.3% (42). The prevalence was higher among hospitalized children, reaching 50.4%. In addition, ESBL colonization was significantly more frequent in children who were HIV-positive (89.7%) than in their counterparts(16.9%).

A survey conducted in Spain reported that *Escherichia coli* was the predominant ESBL-producing organism isolated at hospital admission, accounting for 77.7% of cases, followed by *Klebsiella pneumoniae* at 20.6%. Other ESBL-PE members of the *Enterobacterales* were detected much less frequently, including *Enterobacter cloacae* (0.59%), *Citrobacter freundii* (0.36%), *Enterobacter aerogenes* (0.24%), and *Klebsiella oxytoca* (0.12%).Among ESBL-producing *E. coli* isolates, 1.83% were co-producers of ESBL and carbapenemases, while a substantially higher proportion was observed among ESBL-producing *K. pneumoniae*, where 43.1% (n = 75) also produced carbapenemases(43). Similarly, a study from Northern Cyprus

reported that out of 500 volunteers, 21.4% were colonized with ESBL-producing *Enterobacterales*. Most of the isolates were *Escherichia coli*(94.4%), while *Klebsiellapneumoniae* (2.8%), *Enterobacter cloacae* (0.9%), *Enterobacter aerogenes* (0.9%), and *Providencia rettgeri* (0.9%) were detected at lower frequencies. In addition, 6 participants (1.2%) were colonized with carbapenemase-producing*Enterobacterales* (CPE), with the most common species being *E. coli* (33.3%) and *K. pneumoniae* (33.3%), followed by*E. aerogenes*(16.7%) and K. *oxytoca* (16.7%) (44).

An investigationfrom Hungary compared fecal colonization rates of ESBL-PE among medical students, hospitalized patients and outpatients. Prevalence of colonization among hospitalized patients was 7.4%, compared with 3.1% among outpatients and 2.6% among medical students. The carriage rate of ESBL producers was significantly higher in adults (12 %) than in patients from the pediatric ward (4.1%) ($p< 0.001$). Classifying by hospital ward, the largest incidence was observed among patients inrehabilitation (27.1%), then patients from the intensive care unit (9.3%), and the smallest incidence was observed in non–intensive care wards (6.1 %) (45).

A recent study conducted in Egypt and Saudi Arabia reported Modified Hodge Test (MHT) positivity rates of 43% and 49%, respectively (46,47).Similarly, a study in Iran involving 100 patients found that 50% were carriers of ESBL-PE. The most frequently isolated organism was *Escherichia coli* (85%). No significant association was observed between fecal colonization with ESBL-PE and age ($p = 0.68$), sex ($p = 0.49$), time of diagnosis ($p = 0.21$), antibiotic use within the previous three months ($p = 0.77$), or history of chemotherapy ($p = 0.49$). However, the type of cancer was identified as a significant risk factor for ESBL-PE fecal colonization among cancer patients (48).

A prospective observational study conducted in Morocco investigated rectal colonization with ESBL-PE and CPE among hospitalized newborns. A total of 169 *Enterobacterales* isolates were recovered from rectal screening cultures of 164 neonates. The most frequently identified species were *K. pneumoniae* (83) and *E.coli* (67). Among the total isolates, 58.0% were ESBL producers, including *K. Pneumonia (*66.3%) and *E. coli* (19.4%). Lower frequencies were observed for *Enterobacter cloacae*, *Citrobacter freundii*, *Klebsiella oxytoca*, and *Proteus mirabilis*, accounting for 5.1%, 4.1%, 3.1%, and 2.0%, respectively.Multidrug resistance was present in most isolates that produced ESBL. 99.0% of ESBL-PE isolates were resistant to

gentamicin, 60.2% to trimethoprim-sulfamethoxazole, and 3.1% to ertapenem. Additionally, high levels of quinolone resistance were noted, with 58.2% resistant to norfloxacin and ciprofloxacin and 62.2% resistant to nalidixic acid. On the other hand, amikacin and Fosfomycin continued to be effective against all isolates(49).

A study from Ethiopia reported a high (ESBL-PE) carriage rate of 52%. Colonization was highest among neonates (74%) and lowest among adults (46%). ESBL-producing *Escherichia coli* colonization rates were reported as 11% in neonates, 42% in children, and 42% in adults. In contrast, ESBL-producing *Klebsiella pneumoniae* was most frequently isolated among neonates (68%), followed by children (22%) and adults (7%). The overall carriage rate of carbapenemase-producing*Enterobacterales* (CPE) was 2% (26).Another study conducted at Arba Minch Hospital,Ethiopia, indicates colonization rates of 33.25% for ESBL-PE*Enterobacterales*& 1.43% for carbapenemase-producing *Enterobacterales*. The predominant ESBL producers were *Escherichia coli* (42.46%) and *Klebsiella pneumoniae* (41.09%)(50). In addition, a study from Gondar reported an ESBL-PE colonization rate of 19.9% among HIV-positive children, with all ESBL-producing isolates identified as either *Escherichia coli* (16.2%) or *Klebsiella pneumoniae* (3.7%)(43).

Medboua-Benbalagh*et al.* (2017)(51) reported that 54% of pediatric oncology patients in Algeria carried ESBL-producing *Enterobacterales*, with hematological malignancies strongly associated with carriage and worse outcomes. Abdallah *et al.* (2017) (52) found 68% ESBL carriage and 0.05% carbapenemase carriage among Egyptian patients with gastrointestinal complaints. Kibwana *et al.* (2020)(53) documented a 59.7% prevalence among hospitalized adults in Tanzania, while Arhoune*et al.,* (2017)(49) highlighted 58% ESBL carriage and 1.8% carbapenemase carriage among Moroccan neonates. Desta *et al.* (2016) (54) reported 52% ESBL carriage in Ethiopia, with the emergence of carbapenem-resistant *K. pneumoniae*.

Ouchar Mahamat *et al.* (2019)(55) observed a 44.5% prevalence in Chad, higher in hospitalized patients (51%) than community volunteers (38%). Chaudhary *et al.* (2018) (56) identified 42.5% ESBL-producing *E. coli* in North India, with co-production of AmpC and carbapenemases. Hazirolan*et al.* (2018)(57) reported 34.3% ESBL carriage in a Turkish community. Aklilu *et al.*(2020)(50) found 34.7% ESBL carriage and 1.43% carbapenemase carriage in hospitalized patients in Arba Minch, Ethiopia. Babu *et al.* (2016) (58) reported 62.7% prevalence among

hospitalized and healthy individuals in India. Ruh*et al.* (2019)(44) documented 21.4% ESBL carriage in Northern Cyprus, while Pérez *et al.* (2019)(59) found 7.69% ESBL carriage in Spain. Askari *et al.* (2021)(48) reported 50% ESBL carriage among cancer patients in Iran.

Several studies identified *Escherichia coli* as the most common ESBL-producing organism, including reports from Mexico(60) and Lebanon(61). In contrast, Medboua-Benbalagh*et al.* (2017)(51) and Chaudhary *et al.* (2018)(56) found *Klebsiella pneumoniae* to be the predominant isolate, suggesting geographic variation in colonizing species.

High resistance rates to non-β-lactam antibiotics such as ciprofloxacin were reported in Chad (55), Burkina Faso(62), Morocco(49), Egypt(52), and Tanzania(53). These findings support the role of mobile genetic elements in spreading multidrug resistance and highlight the challenge of treatment in resource-limited settings.Earlier studies in Ethiopia(54), Zimbabwe(63), and India(58) reported complete susceptibility to carbapenems. However, more recent data indicate increasing resistance. Pérez *et al.* (2019)(59) documented co-expression of ESBL enzyme and OXA-48 carbapenemase in Spain, while Arhoune*et al.* (2017)(49) reported carbapenemase carriage among Moroccan neonates.

Lin *et al.* (2019) highlighted the importance of pharmacist-led consultations in addressing infection risks among patients with malignancies(64).Golzarri*et al.* (2019) demonstrated an association between fecal carriage of ESBL-producing organisms and the development of surgical site infections in cancer patients undergoing gastrointestinal and gynecological procedures (60).Christophyt*et al*. (2017) reported the presence of carbapenem-resistant Gram-negative bacilli within the intestinal flora of cancer patients in Lebanon (61). Similarly, Askari *et al.* (2021) confirmed a high prevalence of ESBL colonization among oncology patients in Iran(48).

2.5 Risk factors for ESBL-PE and CPE colonization in oncology

Several specific risk factors have been identified as contributing to an increased risk of ESBL-producing *Enterobacterales* (ESBL-PE) infections among hospitalized patients, including severe comorbid conditions, prolonged or inappropriate antibiotic use, surgical or other invasive procedures, extended hospital or intensive care unit stays prior to infection, and the use of indwelling instrumentslike catheters (65). Although these factors have been consistently reported

across multiple studies, prior exposure to antibiotics remains the most significant independent predictor of ESBL infection(66).

According to one study, longer durations of antibiotic treatment were significantly associated with the acquisition of ESBL-PE and CPE colonization among patients who converted from carriage of susceptible strains to resistant strains (median: 60 days; range: 19-100 days) compared with those who remained colonized with non-ESBL-producing strains (median: 30.5 days; range: 0-159 days; $p = 0.02$). Similarly, prolonged hospital stay was observed in the resistant group (61 ± 27 days versus 42 ± 32 days; $p = 0.03$) (67).In patients with hematological malignancies, ESBL-PE colonization was also associated with an increased risk of bloodstream infection caused by the same strain (relative risk = 3.4; 95% CI: 1.5-7.8; $p = 0.001$). In addition, colonization was linked to shorter survival time (74 ± 62 vs. 95 ± 83 days; $p < 0.001$), longer hospitalization (64 ± 39 vs. 48 ± 32 days; $p = 0.01$), and higher infection-related costs, including antibiotics, hospitalization, and diagnostic investigations ($\$6528 \pm 4348$ vs. $\$4722 \pm 3173$; $p = 0.01$)(67).

Previous studies in immunosuppressed patients have demonstrated that colonization with ESBL-PE is a significant risk factor for the development of BSIs and is associated with higher mortality compared with non-ESBL-PE colonization(68).The association between colonization and subsequent BSI is largely explained by translocation of intestinal flora during the onset of neutropenia. The increased mortality observed in ESBL-PE colonized patients may be attributed to multiple factors, including the presence of virulence determinants, increased susceptibility leading to prolonged hospitalization, frequent and prolonged antibiotic exposure, higher likelihood of persistent colonization and infection, and delays in initiating effective antimicrobial therapy. In addition, unanticipated resistance patterns may hinder the timely administration of appropriate antibiotics, further contributing to poor outcomes(68).

A Turkish studyidentified recent antibiotic usage, prior hospitalization and surgery, diabetic mellitus, overcrowded household conditions, and advanced age as significant risk factors for ESBL-PE carriage(69).Similarly, a study from Cyprus reported that higher status of socio-economy, exposure to antibiotics, the existence of symptom on GI, and international travel was associated with fecal transmission of ESBL-PE (44). In South Korea, prolonged hospital stay and extended intensive care unit admission were identified as independent risk factors for ESBL-PE

colonization (41). Likewise, an Algerian investigation found that antibiotic usage within the previous three months & the presence of hematological malignancies weresignificantly associated with fecal carriage of ESBL-producing *Enterobacterales* (51).

A point prevalence study conducted in Vietnam identified carbapenem therapy and prolonged hospital stay as independent risk factors for fecal transmission of CPE(70).Similarly, a study in India reported that extended intensive care unit stay, mechanical ventilation, and aminoglycoside use were significant risk factors for CPE transmission (71). Another research from India further demonstrated that the existence of permanent implants and nasogastric tubes were independent predictors of intestinal colonization (72).Findings from a case–control study in China revealed that hospital readmission, intra-hospital bedside transfers, and exposure to invasive procedures were significant independent predictors of carbapenem-resistant *Enterobacterales* carriage (73).In Ethiopia, a study conducted in Arba Minch identified chronic diseases, prolonged hospitalization, post-admission oral antibiotic use, rural residence, and sex as statistically significant factors associated with colonization (50).

3. Objective

3.1 General objective

- To assess the Fecal Carriage of Extended-Spectrum β-Lactamase- and Carbapenemase-producing *Enterobacterales* among oncology patients at St. Paul's Hospital Millennium Medical College, Addis Ababa, Ethiopia

3.2 Specific objectives

- To assess the fecal carriage of ESBL-PE and CPE among cancer patients in the oncology unit at SPHMMC, Addis Ababa, Ethiopia, from January to March 2025.
- To determine the antibiotic susceptibility profiles of ESBL-PE and CPE isolates obtained from cancer patients in the oncology unit at SPHMMC during the study period.
- To identify associated factors for fecal carriage of ESBL-PE and CPE species among cancer patients attending the oncology ward of SPHMMCduring the study period.

4. Hypothesis

Ho: Oncology patients at SPHMMC have a significantly higher fecal carriage rate of ESBL and CPE compared to the general population, likely due to their immunocompromised state and increased exposure to antibiotics during cancer treatment.

5. Methods and Materials

5.1 Study Area

The research was undertaken at St. Paul's Hospital Millennium Medical College (SPHMMC), one of Ethiopia's leading public referral hospitals, located in Addis Ababa. The hospital provides comprehensive clinical care to a large and diverse population. With more than 700 inpatient beds, SPHMMC manages an average of 1,200 emergency and outpatient consultations per day.The oncology unit of SPHMMC currently has 8 Oncologists, 2 medical physicists, 2 nuclear Medicine specialists, 1 radiopharmacologist, and 22 oncology nurses. The center has been providing oncology care to an average of 256 patients per month, with 50 patients per week receiving adult care and 14 receiving pediatric care. The major services provided are new patient evaluation, follow-up chemotherapy administration (outpatient and inpatient level), health education and promotion, and intermittent screening campaigns.

5.2 Study design and period

A cross-sectional study design was implemented at SPHMMC over a three-month period, from January through March 2025.

5.3 Population

5.3.1 Source of population

All oncology patients receiving care at SPHMMC, Addis Ababa, Ethiopia.

5.3.2Study population

The source population consisted of cancer patients admitted to the oncology unit of SPHMMC who satisfied the predefined inclusion criteria throughout the study period.

5.4 Inclusion and exclusion criteria

5.4.1 Inclusion criteria

The study enrolled all patients admitted to the cancer unit at SPHMMC with a diagnosed malignancy, provided that informed consent or assent was obtained from the patients themselves or from their parents or legal guardians when applicable.

5.4.2 Exclusion criteria

Patients who were unable to submit a fecal specimen or who had received antibiotic therapy within the two weeks preceding enrollment were excluded from the study.

5.5Studyvariables

5.5.1 Dependent variable

- Prevalence of Extended-Spectrum β-lactamase-producing*Enterobacterales*from fecal specimens
- Prevalence of carbapenemase-producing *Enterobacterales*from fecal specimens
- Antimicrobial susceptibility patterns

5.5.2 Independent variable

The study variables comprised socio-demographic factors (age, sex, and education level), residence, history of hospitalization, antibiotic use and immunosuppressive treatmentwithin the last three months, type of cancer, cancer stage.comorbidities, neutropenia, ICU admission, invasive procedure, indwelling device, pet at home and eating raw food.

5.6 Sample size determination and sampling technique

5.6.1 Sample size determination

The required sample size was estimated using the single population proportion formula, applying a 95% confidence level, 5% precision, and a 10% non-response adjustment. The calculation was informed by a previously reported prevalence of 84.1% from Jimma Medical Center in Ethiopia (74).

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

$$= \frac{(1.96)^2 * 0.841(1-0.841)}{(0.05)^2} = 205.5$$

Where,

n = Sample size

$Z_{\alpha/2}$ = Z-score corresponding to the desired level of confidence

P = proportion of the target population.

d = Margin of sampling error.

After adding a 10% non-response rate, this study's final minimum sample size was226 cancer patients.

5.6.2 Sampling technique

Consecutive sampling was employed for the recruitment of study participants.

5.7 Data collection method and procedures

5.7.1 Questionnaire and Laboratory Methods

A pre-tested, structured questionnaire was used to collect socio-demographic and clinical information.The collectedinformation included age, sex, level of education, residence, history of prior hospitalization, antibiotic use in the preceding three months, and comorbidities such as diabetes mellitus, chronic heart disease, chronic renal disease, and chronic liver disease. To ensure data quality, the questionnaire was administered by trained data collectors, with close attention to accuracy and completeness.

5.7.2 Sample collection, transportation, and processing

Fecal samples were collected using methods appropriate for the patient’s age, condition, and mobility. For adults and older children, stool was collected directly into a clean, wide-mouth, leak-proof, sterile container. For infants and toddlers, feces were collected from diapers using a sterile spoon or tongue depressor, avoiding contamination with urine. For elderly or bedridden patients, fecal samples were collected using a clean bedpan lined with plastic wrap, and then transferred to a sterile container with a sterile spoon or tongue depressor. Samples were immediately transported to the SPHMMC Microbiology Laboratory for culture. Data collectors were trained in proper collection and transport procedures to ensure sample quality.

5.7.3 Identification of ESBL and carbapenemase-producing*Enterobacterales*

The ChromID ESBL and ChromID CARBA media, once inoculated, were incubated at 37°C for 18–24 hours to allow bacterial growth.Colonies suspected to be targeted *Enterobacterales* were selected based on their morphology and pigment production on the ChromID agar. Further identification was performed using conventional biochemical tests, including colony morphology, Citrate utilization, triple Sugar Iron (TSI) agar, urease test, Motility-Indole-Sulfide (MIS) test, and Lysine Iron Agar (LIA). Confirmed *Enterobacterales* isolates were stored at –80°C for further analysis. Positive and negative control strains were used to ensure the accuracy of the screening and identification procedures.

5.7.4 Phenotypic Confirmation of ESBL production

Initial screening for ESBL-producing *Enterobacterales* was performed using ChromID ESBL agar, followed by confirmation using the combination disk technique (CDT). A standardized 0.5 McFarland bacterial suspension was inoculated onto Mueller–Hinton agar (Liofilchem® s.r.l., Italy), after which ceftazidime (30 µg) and cefotaxime (30 µg), both alone and combined with clavulanic acid (30 µg/10 µg), were placed 25 mm apart (center to center). Following incubation at 37°C for 18–24 hours under aerobic conditions, isolates exhibiting an increase of ≥5 mm in inhibition zone diameter in the presence of clavulanic acid were confirmed as ESBL producers.

5.7.5Phenotypic Confirmation of CarbapenemaseProduction

Carbapenemase production was phenotypically confirmed using the modified carbapenem inactivation method (mCIM) for isolates exhibiting meropenem resistance screening results (inhibition zone ≤18 mm). A bacterial suspension was prepared by emulsifying a loopful of the isolate in 2 mL of Tryptic Soy Broth, followed by incubation of a meropenem (10 µg) disk in the suspension at 35°C for 4 hours. A standardized 0.5 McFarland suspension of *Escherichia coli* ATCC 25922 was used to prepare a lawn on Mueller–Hinton agar. The incubated disk was then transferred onto the indicator lawn and incubated at 35°C for 18–24 hours. Interpretation followed CLSI criteria, where inhibition zones ≥19 mm indicated absence of carbapenemase activity, while zones of 6–15 mm or growth within a 16–18 mm zone indicated a positive result.

5.7.6 Antimicrobial Susceptibility Testing

Antibiotic susceptibility profiles were determined using the Kirby–Bauer disk diffusion method. Briefly, pure bacterial colonies obtained from sheep blood agar cultures incubated for 18–20 hours were emulsified in sterile saline and standardized to a turbidity equivalent to a 0.5 McFarland standard. The resulting suspension was evenly spread onto Mueller–Hinton agar plates using a sterile cotton swab, and the plates were allowed to dry for 3–15 minutes before the application of antibiotic disks. The antimicrobial agents evaluated were cefotaxime (5 μg), ceftazidime (10 μg), cefepime (30 μg), meropenem (10 μg), amikacin (30 μg), gentamicin (10 μg), ciprofloxacin (5 μg), and piperacillin/tazobactam (30/6 μg). After incubation at 37°C for 16–18 hours, inhibition zone diameters were measured and interpreted in accordance with the Clinical and Laboratory Standards Institute (CLSI) 2023 guidelines. *Escherichia coli* ATCC 25922 was included as the quality control organism.

5.7.7 Data Quality Assurance

Prior to data collection, training on the study protocol and data collection procedures was provided to the data collectors by the principal investigator, and continuous supervision was carried out throughout the study period. To enhance data quality, the questionnaire was translated into the local language and subsequently back-translated into English to ensure consistency and accuracy. A pilot test involving 5% of the calculated sample size was conducted in the adult oncology unit of SPHMMC, and the questionnaire was refined based on the pretest results. Strict adherence to standard bacteriological techniques was maintained throughout all laboratory procedures to ensure the validity and reliability of the findings.

The performance of ChromID ESBL agar was verified using reference strains from the American Type Culture Collection (ATCC), including ESBL-producing *Escherichia coli* CIP 103982, which produced small reddish colonies; ESBL-producing *Klebsiella pneumoniae* ATCC 700603, characterized by metallic blue colonies; and *Enterococcus faecalis* ATCC 29212, which exhibited no growth. Quality control for culture and antimicrobial susceptibility testing was performed using *E. coli* ATCC 25922, *Staphylococcus aureus* ATCC 25923, and *Proteus mirabilis* ATCC 35659. Furthermore, *K. pneumoniae* ATCC 700603 and *E. coli* ATCC 25922 served as positive and negative controls, respectively, for ESBL detection.

5.7.8 Data processing and analysis

Data were entered through Kobo Toolbox and analyzed using SPSS version 27. Descriptive and bivariate statistical analyses were performed to summarize the data and evaluate associations between variables. Factors with a p-value below 0.25 in the bivariate analysis were included in a multivariable logistic regression model. Adjusted odds ratios (AORs) with 95% confidence intervals (CIs) were calculated to measure the strength of associations, and a p-value <0.05 was considered statistically significant. The results were presented in tables and figures and interpreted according to key demographic and clinical characteristics.

5.8 Operational definitions

Multidrug-resistant: refers to bacterial isolates resistant to at least one antimicrobial agent in three or more classes of antimicrobials. In this study, it indicates organisms isolated from oncology patients that demonstrate multidrug resistance(75).

5.9Ethical consideration

Ethical approval for the study was obtained from the Departmental Research and Ethics Evaluation Committee of the Department of Medical Laboratory Sciences, College of Health Sciences, Addis Ababa University (Reference No. DRERC/802/24/MLS). Permission to conduct the research was subsequently secured from the administration of St. Paul's Hospital Millennium Medical College (SPHMMC). Before enrollment, the objectives of the study were clearly explained to participants, parents, or caregivers, who were provided with sufficient information regarding the purpose of the research and the importance of their participation (Annex I).Informed written consent (Annex II) was obtained from all study participants or from parents/guardians in the case of children, and assent was obtained from children when appropriate. Confidentiality of all participant information was strictly maintained. During the study period, culture results from fecal samples were communicated to the attending physicians or healthcare providers to ensure appropriate patient management.

5.10 Dissemination of findings

The results of this study will be submitted to the Department of Medical Laboratory Sciences, College of Health Sciences, Addis Ababa University, in partial fulfillment of the requirements for the MSc degree and will be presented as part of the thesis defense. The findings will also be communicated to relevant stakeholders, including SPHMMC, to support evidence-based decision-making. Furthermore, the results will be disseminated through presentations at scientific conferences, seminars, and workshops. Efforts will also be made to publish the findings in peer-reviewed scientific journals.

6. Result

6.1 Sociodemographic characteristics of the study participants

A total of 226 oncology patients were enrolled in the study, including 128 (56.6%) females. Most participants lived in urban areas, accounting for 142 (62.8%) of the study population. The mean age was 37.8 years (±20.98 SD), with ages ranging from 1 to 81 years. The majority of participants (89.0%) were younger than 50 years of age (Table 1).

Table 1: Socio-demographic characteristics of study participants in SPHMMC, Addis Ababa, Ethiopia from Jan to Mar 2025

Variables	Category	Frequency (%)
Gender	Male	98 (43.4%)
	Female	128 (56.6%)
Age(years)	Mean (SD)	38.17 (20.8)
Place of residence	Urban	142 (62.8%)
	Rural	83 (36.7%)
Education status	No formal education	87 (38.5%)
	Primary education	86 (38.1%)
	Secondary education	19 (8.4%)
	College and above	34 (15.0%)

6.2. Clinical Profile of Oncology Patients

Among the total cancer patients included in the study, 193 (85.4) had one or more comorbidities, and 194 (85.8) had a history of antibiotic use. Neutropenia was observed in 179 (79.2) patients, and 194 (85.8%) were receiving immunosuppressive treatment. More than half of the patients, 126 (55.8%), had undergone invasive procedures, and 180(79.6) reported a history of

hospitalization within the previous 12 months. Carcinoma was the most common cancer type, accounting for 120 (53.1%) of cases.

Table 2: Clinical profiles of study participants in SPHMMC, Addis Ababa, Ethiopia from Jan to Mar 2025

Clinical variables	Categories	Frequency (%)
Co-morbidities	Yes	193 (85.4)
	No	33 (14.6)
Hospitalization history	Yes	180 (79.6)
	No	46 (20.4)
Previous antibiotic history	Yes	194 (85.8)
	No	32 (14.2)
Type of cancer	Carcinoma	120 (53.1)
	Leukemia	52 (23.0)
	Sarcoma	12 (5.3)
	Myeloma	2 (0.8)
	Lymphoma	2 (0.8)
	Colorectal/Adenosquamos carcinoma	19 (8.4)
	Germcell	5(2.2)
	Neuroectodermal	4 (1.7)
	Melanoma	2 (0.8)
	Other rare	8 (3.5)
Cancer stage	Stage 1	49 (21.7)
	Stage 2	49 (21.7)
	Stage 3	53 (23.5)
	Stage 4	75 (33.2)
Neutropenia	Yes	179 (79.2)
	No	47 (20.8)
ICU admission	Yes	5 (2.2)
	No	221 (97.8)

Invasive procedure	Yes	126 (55.8)
	No	100 (44.2)
Immunosuppressive treatment	Yes	194 (85.8)
	No	32 (14.2)
Indwelling device	Yes	112 (49.6)
	No	114 (50.4)
Pet at home	Yes	80 (35.4)
	No	146 (64.6)
Eat raw food	Yes	72 (31.9)
	No	154 (68.1)

6.3. Magnitude of ESBL-Producing *Enterobacterales*

A total of 226 oncology patients were screened for intestinal carriage of Extended-Spectrum β-lactamase (ESBL)-producing *Enterobacterales* using ESBL CHROMagar. Of these, 162 (71.7%) showed growth of presumptive ESBL-producing isolates, whereas 64 (28.3%) demonstrated no growth. From the culture-positive patients, a total of 179 presumptive ESBL-producing *Enterobacterales* isolates were recovered. 145 (89.5%) patients harbored a single isolate, and 17 (10.5%) of patients carried two different *Enterobacterales*. Phenotypic confirmation using the combined disk test (CDT) verified ESBL production in 158 of the 179 isolates (88.2%). Among the confirmed ESBL-producing *Enterobacterales*, five different bacterial species were identified. *Escherichia coli* was the predominant species (n = 131, 82.9%), followed by *Klebsiella oxytoca* (n = 12, 9.4%) and *Klebsiella pneumoniae* (n = 8, 5.0%). Overall, the magnitude of intestinal carriage of ESBL-producing *Enterobacterales*among the study population was 67.3% (152/226).

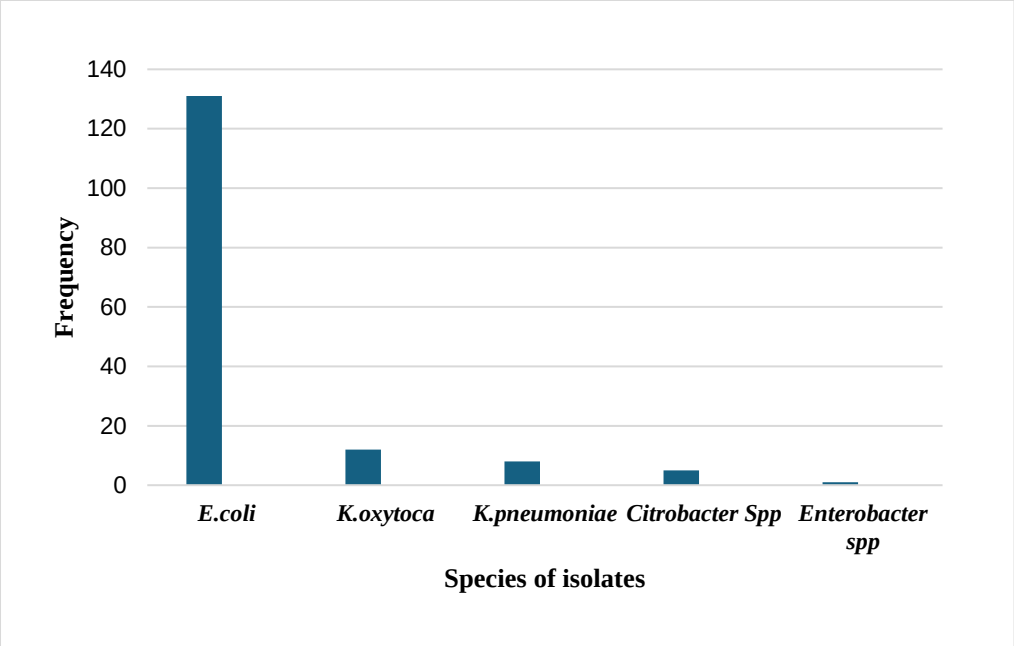


Figure 1: Frequency of ESBL–producing *Enterobacterales* isolated from cancer patients in SPHMMC, Addis Ababa, Ethiopia from Jan to Mar 2025

6.4. Magnitude of Carbapenemase-Producing *Enterobacterales*

A total of 226 oncology patients were screened for intestinal carriage of Carbapenemase-producing *Enterobacterales* using CPE CHROMagar. Of these, 54 (23.9%)patients showed growth of presumptive carbapenemase-producing isolates. From the culture-positive patients, a total of 59 presumptive *Enterobacterales* isolates were recovered. 5 (9.3%) patients harbored two different *Enterobacterales*,while 49 (90.7%) patients had a single isolate. Phenotypic confirmation using the mCIM verified production of carbapenemase in 45 of the 59 isolates (76.3%). Among the confirmed carbapenemase-producing isolates, five different bacterial species were identified. *Escherichia coli* was the predominant species (n = 30, 66.6%), followed by *Klebsiella pneumoniae* (n = 9, 20.0%) and *Klebsiella oxytoca* (n = 4, 8.8%). Overall, the prevalence of intestinal carriage of carbapenemase-producing *Enterobacterales* among the study population was 17.7% (40/226).

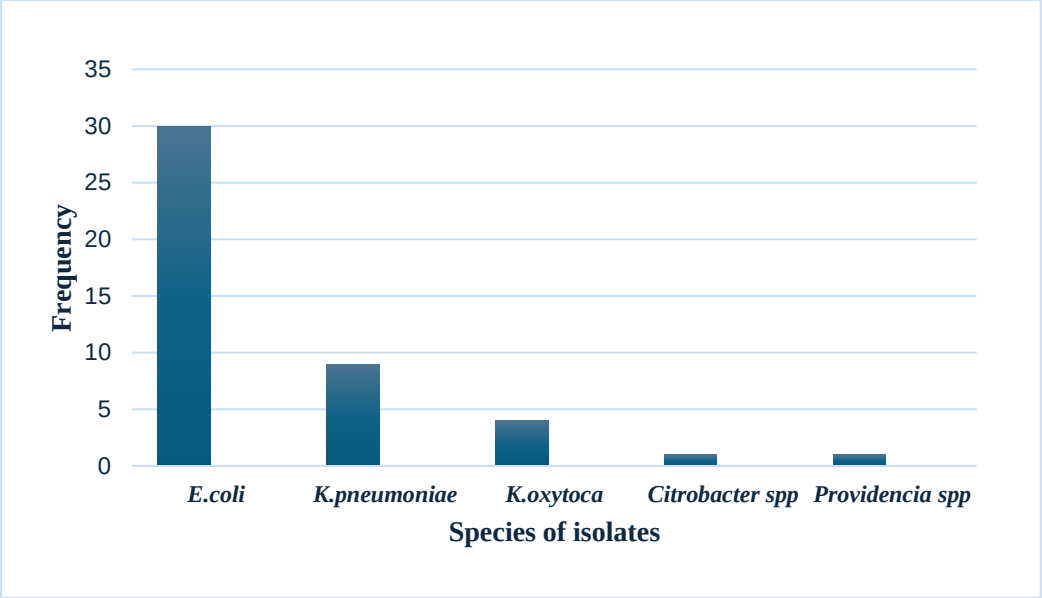


Figure 2: Frequency of Carbapenemase–producing *Enterobacterales* isolated from cancer patients in SPHMMC, Addis Ababa, Ethiopia, from Jan to Mar 2025

6.5Antibiotic Resistance Pattern of ESBL-Producing *Enterobacterales*

Antimicrobial susceptibility tests were performed on 158 ESBL producing*Enterobacterales* High rate of resistance were noted against ciprofloxacin (83, 52.5%) followed by cefepime (68, 43%).While lower resistance rates were noted for gentamicin (29, 18.4%) and meropenem (35, 22.2%). Amikacin showed the lowest resistance rate (7, 4.4%), followed by piperacillin–tazobactam (16, 10.1%). *E. coli* demonstrated particularly high resistance to ciprofloxacin (70, 53.4%) and cefepime (40.5%), whereas resistance to amikacin remained low (6, 4.6%).(Table 4)

Table 3: Antibiotic resistance profiles of ESBL-producing isolates among cancer patients in SPHMMC, Addis Ababa, Ethiopia from Jan to Mar 2025

Isolate (N)	AK n (%)	GEN n (%)	FEP n (%)	CIP n (%)	TZP n (%)	MEM n (%)
E. coli (131)	6(4.6)	21(16)	53(40.5)	70(53.4)	12(9.2)	26(19.8)
K. Oxytoca (12)	1(8.3)	5(41.7)	7(58.3)	6(50)	3(25)	4(33.3)
K. pneumoniae (8)	0(0)	3(37.5)	4(50)	4(50)	1(12.5)	4(50)

Citrobacter spps (5)	0(0)	0(0)	3(60)	3(60)	0(0)	1(20)
Enterobacter spps (2)	0(0)	0(0)	1(50)	0(0)	0(0)	0(0)
Total (158)	7(4.4)	29(18.4)	68(43)	83(52.5)	16(10.1)	35(22.2)

Note: AK: amikacin; GEN: gentamicin; FEP: cefepime; CIP: ciprofloxacin; TZP: piperacillin-tazobactam; MEM: meropenem

6.6Antibiotic Resistance Pattern of Carbapenemase-Producing *Enterobacterales*

Among the 45 carbapenemase producing *Enterobacterales*analyzed, high resistance rates were observed to third- and fourth-generation cephalosporins, with (44, 97.7%) resistance to both ceftazidime (CTZ) and ceftraxone (CTX), and (43, 95.5%) to cefepime (FEP). Ciprofloxacin resistance was also high (42, 93.3%). Amikacin demonstrated comparatively lower resistance at (25, 55.5%). *E. coli* was the predominant isolate and exhibited particularly high resistance to cephalosporins and ciprofloxacin. (Table 5)

Table 4: Antibiotic resistance profiles of CPE-producing isolates among cancer patients in SPHMMC, Addis Ababa, Ethiopia, from Jan to Mar 2025

Isolate (N)	**AK n (%)**	**GEN n (%)**	**CTZ n (%)**	**CTX n (%)**	**FEP n (%)**	**CIP n (%)**	**TZP n (%)**
E. coli (30)	1(56.6)	27(90)	29(96.6)	29(96.6)	28(63.3)	29(96.6)	23(76.6)
K.oxytoca (4)	2(50)	3(75)	4(100)	4(100)	4(25)	3(75)	2(50)
K. pneumoniae (9)	6(66.6)	8(88.8)	9(100)	9(100)	9(55.6)	8(88.9)	5(55.5)
Citrobacter spps (1)	0(0)	0(0)	1(100)	1(100)	1(100)	1(100)	0(0)
Providenciaspps(1)	(0)	0(0)	1(100)	1(100)	1(100)	1(100)	1(100)
Total (45)	25(55.5)	38(84.4)	44(97.7)	44(97.7)	43(95.5)	42(93.3)	31(68.8)

Abbreviations: AK: amikacin; GEN: gentamicin; CTZ: ceftazidime; CTX: cefotaxime; FEP: cefepime; CIP: ciprofloxacin; TZP: piperacillin-tazobactam; MEM: meropenem

6.7Multi-drug resistance pattern of isolates

Among the 158 ESBL-producing *Enterobacterales* isolates, 23 (14.6%) were multidrug-resistant,with *E. coli* accounting for the majority of MDR isolates (18/23). (Table 5)

Table 5: Multidrug resistance level of ESBL-PE isolates from cancer patients in SPHMMC, Addis Ababa, Ethiopia from Jan to Mar 2025

Organism	R0 n (%)	R1 n (%)	R2 n (%)	R3 n (%)	R4 n (%)	R5 n (%)	MDR n (%)
***Citrobacter* spp. (5)**	1(20)	1(20)	3 (60)	0 (0)	0 (0)	0 (0)	0 (0)
***E. coli*(131)**	29(22.1)	58 (44.3)	26 (19.8)	15 (11.5)	3 (2.3)	0 (0)	18 (13.7)
***Enterobacter* spp. (2)**	1 (50)	1 (50)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)
***K. oxytoca*(12)**	3 (25)	2 (16.7)	4 (33.3)	1 (8.3)	2 (16.7)	0 (0)	3 (25)
***K. pneumoniae*(8)**	2 (25)	2 (25)	2 (25)	1 (12.5)	1 (12.5)	0 (0)	2 (25)
Total (158)	36 (22.8)	64 (40.5)	35 (22.2)	17 (10.8)	6 (3.8)	0 (0)	**23 (14.6)**

Of the 45 carbapenemase-producing *Enterobacterales* isolates, 26 (57.8%) were MDR. *E. coli* showed the highest MDR prevalence (66.7%), followed by *K. pneumoniae* (55.6%). (Table 6)

Table 6: Multidrug resistance level of CPE isolates from cancer patients in SPHMMC, Addis Ababa, Ethiopia from Jan to Mar 2025

Organism	R0 n (%)	R1 n (%)	R2 n (%)	R3 n (%)	R4 n (%)	R5 n (%)	MDR n (%)
***Citrobacter* (1)**	0 (0.0)	0 (0.0)	1 (100.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
***E. coli* (30)**	0 (0.0)	4 (13.3)	6 (20.0)	12 (40.0)	8 (26.7)	0 (0.0)	20 (66.7)
***K. oxytoca* (4)**	0 (0.0)	1 (25.0)	2 (50.0)	1 (25.0)	0 (0.0)	0 (0.0)	1 (25.0)
***K. pneumoniae* (9)**	0 (0.0)	1 (11.1)	3 (33.3)	3 (33.3)	2 (22.2)	0 (0.0)	5 (55.6)
***Providencia* (1)**	0 (0.0)	0 (0.0)	1 (100.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
Total (45)	0 (0.0)	6 (13.3)	13 (28.9)	16 (35.6)	10 (22.2)	0 (0.0)	**26 (57.8)**

Note: R0: resistance to no antibiotics, R1-5: resistance to 1, 2, 3, 4, and 5 antibiotics; ≥R3: resistance to at least one agent in 3 or more antimicrobial classes, MDR: multidrug resistance

Antimicrobial classes used to define MDR:Penicillins + β-lactamase inhibitor (Piperacillin-Tazobactam), Cephalosporin (Ceftazidime, cefotaxime, Cefepime), Carbapenem (Meropenem), Aminoglycoside (Amikacin, Gentamicin), Fluoroquinolone (Ciprofloxacin).

6.8 Factors associated with fecal carriage of ESBL-PE among cancer patients

The multivariable analysis showed that repeated antibiotic treatment(AOR=0.38, 95% CI: 0.15–0.95; p=0.04)and indwelling devices(AOR=0.10, 95% CI: 0.05–0.22; p<0.001)were independently associated withCarriage of ESBL-PE among Oncology Patients. Age, sex, residence, and history of hospitalization were not significantly associated after adjustment.

Table 7: Factors associated with fecal carriage of ESBL-PE among cancer patients in SPHMMC, Addis Ababa, Ethiopia from Jan to Mar 2025

Variable	Category	ESBL-negative n (%)	ESBL-positive n (%)	COR (95% CI)	p-value	AOR (95% CI)	p-value
Age	**mean (SD)**	42.77 (19.43)	35.93 (21.14)	0.98 (0.97–1.00)	0.02	0.99 (0.98–1.01)	0.40
Sex	Male	26 (35.1)	72 (47.4)	0.60 (0.34–1.07)	0.08	0.68 (0.34–1.37)	0.28
	Female	48 (64.9)	80 (52.6)	Ref.	—	Ref.	—
Residence	Urban	50 (68.5)	92 (60.5)	1.42 (0.79–2.56)	0.25	1.12 (0.54–2.30)	0.76
	Rural	23 (31.5)	60 (39.5)	Ref.	—	Ref.	—
Repeated antibiotic treatment	Yes	53 (71.6)	141 (92.8)	0.20 (0.09–0.44)	<0.001	0.38 (0.15–0.95)	**0.04***
	No	21 (28.4)	11 (7.2)	Ref.	—	Ref.	—
Indwelling devices	Yes	10 (13.5)	102 (67.1)	0.08 (0.04–0.16)	<0.001	0.10 (0.05–0.22)	**<0.001***
	No	64 (86.5)	50 (32.9)	Ref.	—	Ref.	—
History of hospitalization	Yes	54 (73.0)	126 (82.9)	0.56 (0.29–1.08)	0.08	0.71 (0.32–1.60)	0.41
	No	20 (27.0)	26 (17.1)	Ref.	—	Ref.	—

Note: COR = crude odds ratio; AOR = adjusted odds ratio; CI = confidence interval; SD = standard deviation; Ref. = reference category

6.9. Factors associated with fecal carriage of CPE among cancer patients

Carriage of CPE among oncology patients was independently associated with age (AOR=0.98, 95% CI: 0.96–1.00; *p*=0.03), male sex (AOR=0.37, 95% CI: 0.16–0.86; *p*=0.02), and comorbidities (AOR=9.45, 95% CI: 3.13–28.58; *p*<0.001). In contrast, indwelling devices, history of hospitalization, and regular consumption of raw or undercooked foods were not significantly associated with CPE carriage after adjustment (*p*>0.05).

Table 8: Factors associated with carriage of CPE among cancer patients at SPHMMC, Addis Ababa, Ethiopia from Jan to Mar 2025

Variable	Category	CPE-negative n (%)	CPE-positive n (%)	COR (95% CI)	p-value	AOR (95% CI)	p-value
Age	**mean (SD)**	40.60 (19.25)	26.88 (24.07)	0.97 (0.95–0.99)	<0.001	0.98 (0.96–1.00)	**0.03***
Sex	Male	71 (38.2)	27 (67.5)	0.30 (0.14–0.61)	0.001	0.37 (0.16–0.86)	**0.02***
	Female	115 (61.8)	13 (32.5)	Ref.	—	Ref.	—
Comorbidities	Yes	171 (91.9)	22 (55.0)	9.33 (4.12–21.10)	<0.001	9.45 (3.13–28.58)	**<0.001***
	No	15 (8.1)	18 (45.0)	Ref.	—	Ref.	—
Indwelling devices	Yes	86 (46.2)	26 (65.0)	0.46 (0.23–0.94)	0.03	0.47 (0.20–1.13)	0.09
	No	100 (53.8)	14 (35.0)	Ref.	—	Ref.	—
History of hospitalization	Yes	153 (82.3)	27 (67.5)	2.23 (1.04–4.78)	0.04	1.00 (0.32–3.17)	0.99
	No	33 (17.7)	13 (32.5)	Ref.	—	Ref.	—
Eat raw foods	Yes	64 (34.4)	8 (20.0)	2.10 (0.91–4.82)	0.08	2.08 (0.75–5.77)	0.16
	No	122 (65.6)	32 (80.0)	Ref.	—	Ref.	

Note: COR = crude odds ratio; AOR = adjusted odds ratio; CI = confidence interval; SD = standard deviation; Ref. = reference category.

7. Discussion

Gastrointestinal colonization with extended-spectrum β-lactamase-producing *Enterobacterales* (ESBL-PE) and carbapenemase-producing *Enterobacterales* (CPE) has develop an increasing global public health concern and represents a serious threat, particularly among hospitalized patients. Such colonization has been associated with an increased risk of subsequent infection and contributes to patient-to-patient transmission (51). Gastrointestinal tract colonization is especially concerning in oncology patients due to the effects of cytotoxic chemotherapy and the frequent use of broad-spectrum antibiotics, which disrupt the normal gut microbiota. ESBL- and carbapenemase-producing *Enterobacterales* residing in the gut microbiota act as reservoirs of resistance genes that can be transferred to disease-causing bacterial strains(64).

Over the past decade, the prevalence of ESBL-PE and CPE has increased dramatically worldwide. In Ethiopia, particularly among cancer patients, there has been limited information regarding fecal carriage of these resistant organisms. As far as we are aware, this study is the second to report an alarmingly high prevalence of gastrointestinal colonization with ESBL-PE and CPE among oncology patients. In the present study, most ESBL-PE isolates were recovered from patients with carcinoma (60.3%) compared to other types of malignancy. This finding differs from those reported in studies conducted in Jimma, Ethiopia (89.5%)(74)&and Algeria (73%)(51)where majority of ESBL-producing *Enterobacterales* isolates were obtained from patients diagnosed with hematological malignancies. This may be related to the immunocompromised status of these patients, particularly during neutropenia, and their frequent exposure to antimicrobial agents.

The overall incidence of ESBL-PE fecal carriage in our investigation was 67.3% (95% CI: 61.2–73.4), which is comparable with studies done in Algeria (54%)(51), Egypt (68%)(52) Tanzania (59.7%)(53), Morocco (58%)(49), Ethiopia (52%)(54). The prevalence identified in this study was greater than that reported in a study carried out in Chad (44.5%)(55), India (42.5%)(56), Turkey (34.5)(57),Ethiopia (34.7%)(50), India (62.7)(58), North Cyprus (21.4%)(44), Spain (7.69%)(59),Iran (50%)(48).These differences may be attributed to variations in study populations, inappropriate antibiotic use, differences in infection prevention practices, and variations in detection methods.However, the finding was lower than study reported from Jimma (84.1%)(74) This variation may be associated with excessive use of β-lactam antibiotics,

repeated antibiotic exposure, and cross-transmission in oncology units. Previous studies have demonstrated that repeated antibiotic exposure promotes the development and acquisition of resistant organisms.

The majority of ESBL-producing isolates recovered in this study were identified as *Escherichia coli*followed by *K. oxytoca*. This finding is consistent with reports from Jimma, Ethiopia(74), Mexico(60), and Lebanon(61), which identified ESBL-producing *E. coli* as the predominant colonizing organism. However, other studies conducted in Algeria(51)reported *Klebsiella pneumoniae* as the most frequent isolate. These differences may be related to geographic and environmental variations.

In addition, ESBL-producing *Enterobacterales* demonstrated high resistance rates to multiple non-β-lactam antibiotics, including ciprofloxacin. Comparable report was observed in studies conducted in Jimma, Ethiopia(74), Chad(55), Burkina Faso(62), Ethiopia(50), Morocco(49), Egypt(52), and Tanzania(53). This finding may be due to the co-existence of ESBL genes with other antimicrobial resistance genes on transferable mobile genetic elements, which enables the simultaneous dissemination of resistance to multiple antibiotic classes and leads to multidrug-resistant phenotypes. Inappropriate antibiotic use and lack of susceptibility-guided treatment further promote the selection and spread of resistant strains.

Alarmingly, increased resistance to carbapenems was observed among ESBL-producing isolates. This finding contrasts with previous studies conducted in Addis Ababa, Ethiopia(54), Zimbabwe(63), and India(58), which reported complete susceptibility to carbapenems. This increased resistance may be due to the widespread and inappropriate usage of carbapenems in healthcare settings and increased antibiotic use during the COVID-19 pandemic.

The rate of carbapenemase-producing *Enterobacterales* colonization identified in this study (17.7%) closely aligns with the prevalence reported from India (14.8%)(58) and Jimma (11.5%)(74) but higher than reports from Arba Minch, Ethiopia (1.43%)(50), Morocco (1.8%)(49), Egypt (0.05%)(52). The observed variations may be explained by differences in diagnostic techniques, sample size, study populations, and antimicrobial usage patterns. In this study, carbapenemase production was predominantly detected in *E. coli*, which aligns with

findings from Jimma (74), whereas other studies reported *Klebsiella pneumoniae* as the predominant organism (50,52).

The high rate of ESBL-PE and CPE colonization observed in this study poses a significant risk for hospital-acquired infections and contributes to the spread of antimicrobial resistance in healthcare settings and the community, potentially leading to increased morbidity and mortality.

In the current study, the prevalence of multidrug resistance among ESBL-producing *Enterobacterales* isolates in cancer patients was 14.6%, whereas a substantially higher prevalence (57.8%) was noted among carbapenemase-producing *Enterobacterales*.The markedly elevated MDR level among CPE isolates observed in our study may indicate the accumulation of multiple resistance mechanisms in addition to carbapenemase production, thereby limiting available therapeutic options. The high burden of MDR observed in our isolates is consistent with studies from other low- and middle-income countries that report widespread multidrug resistance among hospitalized patients (76). For instance, astudy conducted in Jimma medical center, Ethiopia report 87.5% ESBL-PE isolates from oncology patients were MDR (77). A recent systematic review and meta-analysis conducted in Ethiopia spanning 15 studies also indicated that the pooled estimate of MDR among extended-spectrum β-lactamase producers was 71.7% (74). Similarly, a study from Iran reported an even higher MDR rate (100%) among ESBL-PE isolates recovered from bloodstream infections in cancer patients (78). The observed variations across studies may be explained by differences in study populations, healthcare settings, antimicrobial usage practices, infection prevention measures, and laboratory methods used for resistance detection. Nevertheless, the increasing burden of MDR among *Enterobacterales* poses a serious challenge for the management of infections in immunocompromised patients such as individuals with cancer.

We also investigated factors associated with ESBL-PE carriage among cancer patients. The multivariable analysis showed that repeated antibiotic treatment was independently associated with ESBL-PE carriage (AOR=0.38, 95% CI: 0.15–0.95; p=0.04). This finding indicates that patients with repeated antibiotic exposure had significantly different odds of ESBL-PE carriage compared with those without repeated treatment. Frequent antibiotic exposure may alter the normal intestinal microbiota and create selective pressure that favors the persistence or acquisition of antimicrobial-resistant organisms. Indwelling devices were also independently associated with ESBL-PE carriage (AOR=0.10, 95% CI:

0.05–0.22; p<0.001). Indwelling devices can provide opportunities for bacterial colonization and transmission, particularly among hospitalized and immunocompromised patients.

Carriage of CPE among oncology patients was independently associated with age, male sex, and comorbidities. Age was independently associated with CPE carriage (AOR=0.98, 95% CI: 0.96–1.00; p=0.03). This suggests that each one-year increase in age was associated with approximately a 2% change in the odds of CPE carriage. The association may be related to differences in healthcare utilization, cumulative antimicrobial exposure, and underlying health conditions across age groups. A significantly different odds of CPE carriage between males and females were also observed in which male sex was independently associated with CPE carriage (AOR=0.37, 95% CI: 0.16–0.86; p=0.02). Differences in healthcare exposure, antimicrobial use, comorbidities, or other unmeasured behavioral and clinical factors may partly explain this finding. Comorbidities showed a strong independent association with CPE carriage (AOR=9.45, 95% CI: 3.13–28.58; p<0.001). Patients with comorbid conditions had approximately nine-fold higher odds of CPE carriage compared with those without comorbidities. This may be explained by greater healthcare utilization, repeated antimicrobial exposure, prolonged hospital contact, and increased susceptibility to colonization among patients with underlying illnesses(79).

8. Conclusion

Gastrointestinal carriage of ESBL- and carbapenemase-producing *Enterobacterales* is prevalent among oncology patients, with *E. coli* representing the leading isolate and *Klebsiella* spp. occurring subsequently.Colonizing strains exhibited resistance to multiple antibiotic classes, including fluoroquinolones and cephalosporins, and some showed reduced susceptibility to carbapenems. Patients with carcinoma appeared particularly vulnerable to multidrug-resistant intestinal colonization, highlighting the risk posed by both solid and haematological malignancies in the context of chemotherapy and frequent antibiotic exposure.

These findings highlight the urgent importance of strengthening infection prevention and antimicrobial stewardship programs in oncology units. Effective reduction of transmission and prevention of infections require routine surveillance for resistant *Enterobacterales*, judicious use of broad-spectrum antimicrobials, and rigorous compliance with infection control and hygiene practices.Addressing these challenges is essential for protecting immunocompromised patients, decreasing morbidity, and mitigating the wider public health impact of antimicrobial resistance.

9. Recommendations

- Oncology units should implement regular screening for gastrointestinal colonization with ESBL and Carbapenemase-producing *Enterobacterales* to identify high-risk patients early.
- Judicious use of broad-spectrum antibiotics is essential to prevent the selection and spread of multidrug-resistant organisms. Antibiotic therapy should be guided by susceptibility testing whenever possible.
- Larger, multicentre studies are needed to explore risk factors for colonization, the dynamics of resistance gene transfer, and effective interventions to reduce colonization and infection rates.

10. Strengthsand Limitations

10.1 Strength of the study

- To the best of our knowledge, limited studies of this kind have been conducted in Ethiopia to investigate fecal carriage of ESBL- and carbapenemase-producing *Enterobacterales* among oncology patients, generating important baseline information for antimicrobial resistance surveillance and infection prevention and control strategies.

10.2 Limitations of the study

- This study was conducted at a single center, which may limit the generalizability of the findings to other regions or healthcare settings.
- The sample size for certain cancer subtypes and rare bacterial isolates was small, reducing statistical power for subgroup analyses.

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Annex

Annex 1: Information Sheet for Oncology Patients

Annex 1.1: Information Sheet for Oncology Patients (English Version)

Introduction:

You are kindly invited to participate in a research study being conducted at St. Paul's Millennium Medical College Hospital. The purpose of this study is to investigate the prevalence of fecal carriage of drug-resistant bacteria, specifically Extended Spectrum β-Lactamase (ESBL) and Carbapenemase-producing *Enterobacterales*, among oncology patients. This research aims to better understand the presence and spread of these resistant bacteria, which pose significant treatment challenges due to limited antibiotic options.

Purpose:

The primary goal of this study is to determine the prevalence of ESBL and Carbapenemase-producing *Enterobacterales* in fecal samples of oncology patients. By identifying the carriers of these drug-resistant bacteria, we hope to guide infection control measures and provide valuable information for future treatment strategies.

Duration:

The study will last for approximately five months, depending on the availability of participants. However, your participationrequire only a one-time fecal sample collection.

Procedure to be carried out:

If you agree to participate, a small fecal sample was collected using a sterile swab. This sample was analyzed in the microbiology laboratory to detect the presence of ESBL and Carbapenemase-producing bacteria. The procedure is straightforward and was carried out by trained principal investigator.

Risk and Discomfort:

There are minimal risks involved in the collection of fecal samples. The process is non-invasive and should not cause any discomfort. All samples were handled with strict confidentiality and were processed following standard safety protocols to ensure privacy.

Expected Benefits:

There are no direct benefits to you for participating in this study. However, the information gathered was help improve infection control practices and treatment approaches for future oncology patients by providing data on the spread of drug-resistant bacteria in this population.

Confidentiality:

Your privacy is highly respected, and all information related to your participation will remain confidential. The collected data was used solely for research purposes. If the study results are published, your name or any personal identifiers will not be disclosed. All collected data was stored securely, either in locked cabinets or password-protected electronic files.

Voluntary Participation and Withdrawal from the Study:

Your participation in this study is entirely voluntary. You have the right to refuse or withdraw from the study at any time without any consequences for your current or future medical care.

Contact Information:

If you have any questions about the study, you can contact:

Principal Investigator: Tsedale Woldu

- Tel: 0913460451
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Advisors:

- Melese Hailu (MSc, PhD), Email: melese.hailu@aau.edu.et
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- MamudeDinkiye (PhD), Email: mdinkiye@gmail.com

Annex 2. Consent and Assent Form

Annex 2.1: Consent Form for Oncology Patients (English Version)

Serial no: _____

I, _____, have been informed about the research study titled "Prevalence of Fecal Carriage Extended Spectrum β-Lactamase and Carbapenemase-Producing *Enterobacterales* among Oncology Patients in St. Paul's Millennium Medical College Hospital, Addis Ababa, Ethiopia." I understand the purpose of the study, the procedures involved, and the potential risks and benefits associated with my participation.

I understand that my participation in this study is voluntary and that I have the right to withdraw at any time without any consequences. I understand that all information collected during the study was kept confidential and used only for research purposes.

I have had the opportunity to ask questions about the study, and all my queries have been answered to my satisfaction. I agree to participate in this research study voluntarily.

Participant's Signature: _____

Date: _____

Researcher's Signature: _____

Date: _____

Annex 2.2: Assent Form for Oncology Patients (English Version)

Serial no: _____

I, _____, have been informed about the research study titled "Prevalence of Fecal Carriage Extended Spectrum β-Lactamase and Carbapenemase-Producing *Enterobacterales* Among Oncology Patients in St. Paul's Millennium Medical College Hospital, Addis Ababa, Ethiopia." I understand that I am being asked to participate in this study.

I understand that my participation is voluntary and that I can choose not to participate or withdraw at any time without any consequences. I understand that all information collected during the study was kept confidential.

I have had the opportunity to ask questions about the study, and all my queries have been answered to my satisfaction. I agree to participate in this research study voluntarily.

Parent or Guardian Signature: _____

Date: _____

Annex 2.3: Information Sheet for Oncology Patients (Amharic Version)

አባሪ 1፡ ለኦንኮሎጂታካሚዎችየመረጃወረቀት

አባሪ 1.1፡ ለኦንኮሎጂታካሚዎችየመረጃወረቀት (የአማርኛቅጂ)

መግቢያ:-

በቅዱስጳውሎስሚሊኒየምሜዲካልኮሌጅሆስፒታልእየተካሄደባለውየምርምርጥናትላይእንድትሳተፉበ አክብሮትተጋብዘዋል። የዚህጥናትዓላማየሚቋቋሙባክቴሪያዎችንበተለይም Extended Spectrum β-Lactamase (ESBL) **እና** Carbapenemase-producing *Enterobacterales*, **በኦንኮሎጂህመምተኞችመካከልያለውንየሰገራማንጓዝስርጭትመመርመርነው። ይህጥናትየነዚህንተከላካይባክቴሪያመኖርእናመስፋፋትየበለጠለመረዳትያለመሲሆንይህምበተወሰኑየ አንቲባዮቲክአማራጮችምክንያትከፍተኛየሕክምናፈተናዎችንይፈጥራል።**

ዓላማ:-

የዚህጥናትዋናግብበኦንኮሎጂህመምተኞችሰገራናሙናዎችውስጥ የ ESBL **እና** Carbapenemase-producing *Enterobacterales***ስርጭትንመወሰንነው። የእነዚህመድኃኒቶችንየመቋቋምችሎታያላቸውባክቴሪያዎችተሸካሚዎችንበመለየትየኢንፌክሽንመቆ ጣጠሪያእርምጃዎችንለመምራትእናለወደፊቱየሕክምናስልቶችጠቃሚመረጃንለመስጠትተስፋእናደር ጋለን።**

የሚፈጀውጊዜ:-

ጥናቱበተሳታፊዎችአቅርቦትላይበመመስረትለአምስትወራትያህልይቆያል።
ይሁንእንጂተሳትፎየአንድጊዜየሰገራናሙናመሰብሰብብቻይፈልጋል።

የሚከናወኑሂደቶች፡-

ለመሳተፍከተስማሙትንሽየፈስታልናሙናበንጽሕናስዋብበመጠቀምይሰበሰባል. ይህናሙና ESBL እና Carbapenemase የሚያመነጩባክቴሪያዎችመኖራቸውንለማወቅበማይክሮባዮሎጂቤተሙከራውስጥይተነተናል።
አሰራሩቀላልእናበሰለጠነዋናመርማሪይከናወናል።

ስጋትእናምቾትማጣት;

የሰገራናሙናዎችንበሚሰበስቡበትጊዜአነስተኛአደጋዎችአሉ.
ሂደቱወራሪአይደለምእናምንምአይነትምቾትማምጣትየለበትም.
ሁሉምናሙናዎችበጥብቅሚስጥራዊነትይያዛሉእናግላዊነትንለማረጋገጥመደበኛየደህንነትፕሮቶኮሎ ችንበመከተልይከናወናሉ።

የሚጠበቁጥቅሞች፡-

በዚህጥናትውስጥለመሳተፍምንምአይነትቀጥተኛጥቅሞችየሉም።
ይሁንእንጂየተሰበሰበውመረጃበዚህህዝብውስጥመድሃኒትንየሚቋቋሙባክቴሪያዎችስርጭትላይመረ ጃንበማቅረብየኢንፌክሽንመቆጣጠሪያልምዶችንእናለወደፊቱየኦንኮሎጂታካሚዎችየሕክምናዘዴዎች ንለማሻሻልይረዳል።

ሚስጥራዊነት፡-

የእርስዎግላዊነትበጣምየተከበረነው።
እናከእርስዎተሳትፎጋርየተያያዙሁሉምመረጃዎችሚስጥራዊእንደሆኑይቆያሉ።
የተሰበሰበውመረጃለምርምርዓላማዎችብቻጥቅምላይይውላል። የጥናትውጤቶቹከታተሙ፣ የእርስዎስምወይምማንኛውምየግልመለያአይገለጽም።
ሁሉምየተሰበሰቡመረጃዎችበተቆለፉካቢኔቶችወይምበይለፍቃልበተጠበቁኤሌክትሮኒክስፋይሎችውስ ጥደህንነቱበተጠበቀሁኔታይከማቻሉ።

በፈቃደኝነትተሳትፎእናከጥናቱመውጣት፡-

በዚህጥናትውስጥያለዎትተሳትፎሙሉበሙሉበፈቃደኝነትነው።

ለአሁኑወይምለወደፊትየሕክምናእንክብካቤዎምንምአይነትመዘዝሳይኖርበማንኛውምጊዜጥናቱንየመ
ተውመብትአልዎት።

የእውቂያመረጃ፡-

ስለጥናቱማንኛቸውምጥያቄዎችካሉዎትማነጋገርይችላሉ፡-

ዋናተመራማሪ ፡- ፀዳለወልዱ

• በስልክቁጥር 0913460451

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አማካሪዎች፡-

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Annex 3. Consent and Assent Form (Amharic Version)

አባሪ 2.1፡ለኦንኮሎጂታካሚዎችየስምምነትቅጽ (የአማርኛቅጂ)

መለያቁጥር፡_____

እኔ፣ _____፣ በአዲስአበባ፣ አዲስአበባ፣
ኢትዮጵያበሚገኘውበቅዱስጳውሎስሚሊኒየምሜዲካልኮሌጅሆስፒታልውስጥበሚገኘውኦንኮሎጂታካ
ሚዎችመካከልመድሃኒትንየሚቋቋሙባክቴሪያዎችንበተለይም Extended Spectrum β-Lactamase (ESBL) እና Carbapenemase-producing Enterobacterales, ስርጭት” በሚልርዕስስለተደረገውየምርምርጥናትተነግሮኛል። የጥናቱዓላማ፣ የተካተቱትሂደቶች፣ እናከእኔተሳትፎጋርተያይዘውሊኖሩየሚችሉትንአደጋዎችእናጥቅሞችተረድቻለሁ።

በዚህጥናትውስጥያለኝተሳትፎበፈቃደኝነትእንደሆነእናበማንኛውምጊዜያለምንምመዘዝየማቋረጥመ
ብትእንዳለኝተረድቻለሁ።
በጥናቱወቅትየሚሰበሰቡትመረጃዎችበሙሉሚስጥራዊሆነውለምርምርዓላማዎችብቻእንደሚውሉተ
ረድቻለሁ።

ስለጥናቱጥያቄዎችንለመጠየቅእድሉንአግኝቻለሁ, እናሁሉምጥያቄዎችየእኔንእርካታአግኝተዋል።
በዚህየምርምርጥናትላይበፈቃደኝነትለመሳተፍተስማምቻለሁ።

የተሳታፊፊርማ፡- _____

ቀን፡_____

የተመራማሪፊርማ፡- _____

ቀን፡_____

አባሪ 2.2፡ ለኦንኮሎጂታካሚዎችየድጋፍቅጽ (የአማርኛቅጂ)

መለያቁጥር፡- _____

እኔ _____ “Fecal Carriage Extended Spectrum β-Lactamase and Carbapenemase-Producing Enterobacteralesበቅዱስጳውሎስሚሊኒየምሜዲካልኮሌጅሆስፒታል፣ አዲስአበባ፣ ኢትዮጵያውስጥበሚገኙኦንኮሎጂታካሚዎችመካከልመስፋፋት”
በሚልርዕስስለተደረገውየምርምርጥናትተነግሮኛል።
በዚህጥናትእንድሳተፍእየተጠየቅኩእንደሆነተረድቻለሁ።

የእኔተሳትፎበፈቃደኝነትእንደሆነእናምንምውጤትሳያስከትልበማንኛውምጊዜላለመሳተፍወይምላለመውጣትመምረጥእንደምችልተረድቻለሁ።
በጥናቱወቅትየሚሰበሰቡትመረጃዎችበሙሉበሚስጥርእንደሚቀመጡተረድቻለሁ።

ስለጥናቱ ጥያቄዎችን ለመጠየቅ እድሉን አግኝቻለሁ, እናሁሉምጥያቄዎችየእኔንእርካታአግኝተዋል.
በዚህየምርምርጥናትላይበፈቃደኝነትለመሳተፍተስማምቻለሁ።

የወላጅ ወይም የአሳዳጊ ፊርማ፡ _____

ቀን፡ _____

Annex 3. Questionnaire

Patient code number / Barcode: _____

1.Age (years) _____

2.Gender ☐Male ☐Female

3. Educational Status

☐No formal education ☐Primary ☐Secondary

☐Diploma ☐University degree

4. Residence ☐Urban ☐Rural

5. Patient Setting☐Inpatient ☐Outpatient

6. Has the patient experienced any of the following symptoms? ***(Select all that apply)***

☐Fever ☐Sepsis ☐UTI symptoms ☐Diarrhea

☐Respiratory symptoms ☐Wound infections ☐No symptoms

7. Presence of Any Comorbidities? ***(Select all that apply)***

☐Diabetes ☐Hypertension ☐Kidney disease

☐Cardiovascular disease ☐Immunosuppression

☐Other (please specify): _____

8. Type of Cancer (if applicable)

☐Hematological (Blood cancer) ☐Solid tumor

9. Specific Type of Solid Tumor (if applicable)

☐Breast cancer ☐Gastric cancer ☐Colorectal cancer

☐Cervical cancer ☐Lung cancer

☐Other (specify): _____

10. Stage of Cancer

☐Stage I ☐Stage II ☐Stage III ☐Stage IV

11. Has the Patient Started Cancer Therapy?

☐Yes ☐No

12. Types of Cancer Therapy Received *(Select all that apply)*

☐Chemotherapy ☐Radiotherapy ☐Surgery

☐Immunosuppressive therapy ☐Combined therapy

☐Other (specify): _____

13. History of Hospitalization in the Last 6 Months

☐Yes ☐No

14. History of Repeated Antibiotic Treatment?

☐Yes ☐No

15. Does the Patient Have Neutropenia?

☐Yes ☐No

16. Has the Patient Undergone Any Invasive Procedures (e.g., Surgery, Catheterization, etc.)?

☐Yes ☐No

17. Was the Patient Admitted to an Intensive Care Unit (ICU) During Hospitalization?

☐Yes ☐No

18. Is the Patient Receiving Any Immunosuppressive Treatment (e.g., Corticosteroids, Chemotherapy)?

☐Yes ☐No

19. Does the Patient Have Any Indwelling Devices? *(Select all that apply)*

☐Endotracheal tube ☐Central venous catheter ☐Urinary catheter

☐Surgical site drainage ☐Hemodialysis catheter

☐Other (specify): _____

20. Do You Have Any Pets at Home?

☐Yes ☐No

21. Do You Regularly Consume Raw or Undercooked Foods?

☐Yes ☐No

Questionnaire (Amharic Version)

የታካሚውመለያቁጥር / ባርኮድ: _____

1. **ዕድሜ (በዓመት):** _____

2. **ጾታ☐ወንድ☐ሴት**

3. **የትምህርትደረጃ**

☐መደበኛትምህርትየሌለው☐የመጀመሪያደረጃ☐ሁለተኛደረጃ

☐ዲፕሎማ☐የዩኒቨርሲቲዲግሪ

4. **የመኖሪያአካባቢ☐ከተማ☐ገጠር**

5. **የታካሚውሁኔታ**

☐ተኝቶየሚታከም / አልጋየያዘ☐መጥቶየሚታከም / የውጭታካሚ

6. **ታካሚውከሚከተሉትየህመምምልክቶችመካከልየትኛውንአስተውሏል? (የሚመለከቱትንሁሉይምረጡ)**

☐ትኩሳት☐የደምመመረዝ / ሴፕሲስ☐የሽንትትቦኢንፌክሽንምልክቶች

☐ተቅማጥ☐የመተንፈሻአካላትሕመምምልክቶች

☐የቁስልኢንፌክሽን☐ምንምምልክትአልታየም

7. **ሌላተጓዳኝበሽታ/ሕመምአለባቸው? (የሚመለከቱትንሁሉይምረጡ)**

☐የስኳርበሽታ☐የደምግፊት☐የኩላሊትበሽታ

☐የልብናየደምዝውውርሥርዓትበሽታ☐የበሽታመከላከልአቅምመዳከም

☐ሌላ (እባክዎንይጥቀሱ): _____

8. **የካንሰርዓይነት (የሚመለከተውከሆነ)**

□የደምካንሰር□ኤጢአዊካንሰር

9. የተለየየእጢአዊካንሰርዓይነት (የሚመለከተውከሆነ)

□የጡትካንሰር□የጨጓራካንሰር□የፊንጣጣናየትልቅአንጀትካንሰር

□የማኅጸንበርካንሰር□የሳንባካንሰር

□ሌላ (እባክዎንይጥቀሱ): _____

10. የካንሰርደረጃ

□ደረጃ 1 □ደረጃ 2 □ደረጃ 3 □ደረጃ 4

11. ታካሚውየካንሰርሕክምናጀምሯል?

□አዎ□አልጀመረም

12. የተወሰዱየካንሰርሕክምናዓይነቶች (የሚመለከተውሁሉይምረጡ)

□የኬሚካልሕክምና (ኬሞቴራፒ) □የጨረርሕክምና (ራዲዮቴራፒ)

□ቀዶጥገና□ከሽታመከላከልአቅምንየሚቀንሱሕክምናዎች

□ጥምርሕክምና□ሌላ (እባክዎንይጥቀሱ): _____

13. ባለፉት 6 ወራትውስጥበሆስፒታልተኝተውየታከሙበትታሪክአለ?

□አዎ□የለም

14. በተደጋጋሚየፀረ-ባክቴሪያ (አንቲባዮቲክ) ሕክምናየመውሰድታሪክአለ?

□አዎ□የለም

15. ታካሚውየነጭየደምሴልማነስ (ኒውትሮፔኒያ) አለበት?

□አዎ□የለበትም

16. ታካሚውማንኛውንምዓይነትበሰውነትውስጥየሚገቡየሕክምናሂደቶችን (ለምሳሌቀዶጥገና፣ካቴተር/ቱቦማስገባትወዘተ) አድርጓል?

□አዎ□አላደረገም

17. ታካሚውበሆስፒታልቆይታውወቅትበጽኑሕመማንመከታተያክፍል (አይ.ሲ.ዩ) ገብቶነበር?

□አዎ□አልገባም

18. ታካሚውየበሽታመከላከልአቅምንየሚቀንሱሕክምናዎችን (ለምሳሌኮርቲኮስቴሮይድ፣ኬሞቴራፒ) እየወሰደነው?

☐አዎ☐እየወሰደአይደለም

19. ታካሚውበሰውነቱላይየተገጠሙ/የገቡየሕክምናመሣሪያዎች (ካቴተሮች/ቱቦዎች) አሉት? (የሚመለከተውንሁሉይምረጡ)

☐ያመተንፈሻቱቦ☐ማዕከላዊየደም ሥርካቴተር☐ያሽንትቱቦ / ካቴተር

☐ያቀዶጥገናቦታፈሳሽማውጫቱቦ☐ያኩላሊትእጥበት (ሄሞዲያሊሲስ) ካቴተር

☐ሌላ (እባክዎንይጥቀሱ): _____

20. በቤትዎውስጥየሚያሳድጉትየቤትእንስሳአለዎት?

☐አዎ☐ያለኝም

21. ጥሬወይምበሚገባያልበሰሉምግቦችንአዘውትረውይመገባሉ?

☐አዎ☐አልመገብም

Declaration

The undersigned declares that this thesis complies with the regulations of the University and meets the accepted standards with respect to originality and quality. PI also agrees to accept responsibility for the scientific ethical and technical conduct of the research project and for provision of required progress reports.

M.Sc. candidate: Tsedale Woldu (B.Sc.)

Signature: _____

Date of submission: _____

This thesis has been submitted with our approval as advisors.

Advisor: MeleseHailu(MSc, PhD)

Signature: _____

Date: _____

Place: Addis Ababa, Ethiopia.

Advisor: Dessie Abera (MSc, PhD Candidate)

Signature: _____

Date: _____

Place: Addis Ababa, Ethiopia.