

High Rate of Quinolone, Ampicillin and High-Level Gentamicin Resistance among Enterococcal Isolates Colonizing Patients in Lagos

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ABSTRACT

Enterococci are commensal bacteria that can cause life threatening infections in debilitated and immunocompromised patients. This study was conducted to determine the antimicrobial susceptibility pattern of enterococcal isolates colonizing hospitalized patients in Lagos University Teaching Hospital, Lagos, Nigeria. Ninety-two enterococcal isolates were tested by the E-test MIC method against Vancomycin, Teicoplanin, Ampicillin, Ciprofloxacin and Gentamicin. One percent (1%) were resistant to Vancomycin, 0% to Teicoplanin and Linezolid, 20.7% to Ampicillin, 58.7% to Ciprofloxacin, and 38% exhibited high level resistance to Gentamicin. Antimicrobial stewardship measures are required to preserve the efficacy of currently available antimicrobial agents.

Keywords: *Enterococci, Resistance, Nigeria*

INTRODUCTION

Enterococci are Gram positive coccal bacteria that form part of the normal flora in humans but are also responsible for important infections including urinary tract infections and endocarditis¹. These organisms, which were formerly classified as streptococci, are also a very important cause of hospital acquired infections in severely debilitated and immune compromised patients. Enterococcal infections have come to the fore because of the ability of some strains, which are resistant to multiple antimicrobial agents including glycopeptides, to cause life threatening infections in debilitated and immune compromised patients including patients with hematological malignancies, transplant recipients and intensive care patients. They were the fourth most commonly isolated organism in bacteraemia in the US and eighth in Brazil².

There have been outbreaks of enterococcal infections, especially involving multidrug resistant clones, reported in hospitals and long-term care facilities around the world, and although reports have not been made from Nigeria, it is possible that this may begin to occur in healthcare settings given the improvements in critical care in the country^{3,4}. Only a few

antimicrobial are useful in the management of enterococcal infections and even fewer are accessible in developing countries. Ampicillin-gentamicin combinations are the mainstay for treatment of serious infections due to enterococci and flouroquinolones are an important alternative treatment. Development of widespread resistance to these agents will portend grave consequences in management of enterococcal infections.

Enterococci with multiple drug resistance including glycopeptide resistance have been reported in Nigeria^{5,6}. Monitoring resistance patterns of these organisms will help to guide antibiotic policies and limit the emergence of multi-drug resistant strains. Since the primary habitat of these organisms is the human gastrointestinal tract, from where they spread to cause infections in other organs, stool samples were used to isolate enterococci reported in this study.

MATERIALS AND METHODS

This study was conducted on 92 enterococcal isolates recovered from 73 stool samples collected from 73 in-patients (31 males and 42 females) of the Lagos University Teaching Hospital (LUTH), Nigeria. Seventeen subjects had two enterococcal species isolated from their stool and one subject had two species isolated.

LUTH is a 761-bed tertiary hospital, the largest of three tertiary hospitals in Lagos state and serves Lagos and neighbouring states. These samples were collected as part of a surveillance programme for vancomycin-resistant enterococci. Isolates were recovered using bile-esculin-azide broth and agar (Condalab, Spain),

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and characterized by Gram staining and catalase testing. Enterococci were identified to specie level by Matrix Assisted Laser Desorption Ionization (MALDI) using the VITEK2 MS system (Bio-Me'rieux, France) in the Institute of Infectious disease Epidemiology, University of Leipzig, Germany.

Antimicrobial Susceptibility Testing

Isolates were tested by the agar gradient method (Etest) on Mueller-Hinton agar. The antimicrobial agents tested were Vancomycin, Teicoplanin, Ampicillin, Ciprofloxacin and Gentamicin. CLSI MIC breakpoints⁷ were used to classify the isolates as sensitive, intermediate and resistant. High level gentamicin resistance (HLGR) was determined using a gentamicin E-test strip. An MIC >500µg/ml was designated

HLGR. Quality control for antibiotic strips used was performed using institutional control organisms (*Enterococcus faecalis vanA* and *E. faecalis vanB*).

RESULTS

Of 92 enterococcal isolates included in this study, 41 were *E. faecalis*, 37 were *E. faecium*, 10 were *E. gallinarum*, and one each of *E. avium*, *E. casseliflavus*, *E. durans* and *E. hirae* (Table 1). Overall, 1% were resistant to Vancomycin, 0% to Teicoplanin and Linezolid, 19% to Ampicillin, 54% to Ciprofloxacin, and 35% exhibited high level resistance to Gentamicin (Table 2). Table 3 shows the susceptibility profile of the various species of enterococci to antimicrobial agents.

DISCUSSION

Enterococci have emerged as a leading

Table 1: Species of Enterococci included in this Study

Specie	No.	(%)
<i>E. faecalis</i>	41	(44.56)
<i>E. faecium</i>	37	(40.22)
<i>E. gallinarum</i>	10	(10.86)
<i>E. casseliflavus</i>	1	(1.09)
<i>E. avium</i>	1	(1.09)
<i>E. durans</i>	1	(1.09)
<i>E. hirae</i>	1	(1.09)
Total	92	(100)

Table 2: Susceptibility of Isolates Tested to Selected Antimicrobial Agents

Antibiotics	Susceptibility in no. (%) of strains					
	Susceptible		Intermediate		Resistant	
Vancomycin	82	(89.1)	9	(9.8)	1	(1.1)
Teicoplanin	91	(98.9)	1	(1.1)	0	(0)
Ampicillin	73	(79.3)	0	(0)	19	(20.7)
Ciprofloxacin	14	(15.2)	24	(26.1)	54	(58.7)
Linezolid	70	(78.1)	20	(21.7)	0	(0)
Gentamicin(HLR)	57	(62)	-	-	35	(38)

Table 3: Antimicrobial Susceptibility Pattern of Isolates by Species

	N	Vancomycin			Teicoplanin			Ampicillin			Ciprofloxacin			Linezolid			Gentamicin	
		S	I	R	S	I	R	S	I	R	S	I	R	S	I	R	HLR	I
<i>All species</i>	92	82	9	1	91	1	0	73	0	19	14	24	54	70	20	0	35	57
<i>E. avium</i>	1	-	1	-	1	-	-	1	-	-	-	1	-	1	-	-	-	1
<i>E. casseliflavus</i>	1	-	1	-	1	-	-	1	-	-	-	-	1	1	-	-	-	1
<i>E. durans</i>	1	1	-	-	1	-	-	1	-	-	1	-	-	1	-	-	-	1
<i>E. faecalis</i>	41	41	-	-	41	-	-	40	-	1	5	9	27	31	10	-	22	19
<i>E. faecium</i>	37	33	3	1	36	-	1	19	-	18	4	11	22	27	10	-	11	26
<i>E. gallinarum</i>	10	6	4	-	10	-	-	10	-	-	3	3	4	10	-	-	2	8
<i>E. hirae</i>	1	1	-	-	1	-	-	1	-	-	1	-	-	1	-	-	-	1

cause of hospital acquired infection especially in patients with severe debilitating disease. The gastrointestinal tract being the major reservoir of this organism, it has been recommended that surveillance of fecal specimen for glycopeptide resistance be carried out routinely and periodically⁸. This informed the conduct of this study.

Enterococci are renowned for resistance or relative resistance to several classes of antibiotics leaving few options for treatment of enterococcal infections. Flouroquinolones remain one of the few classes of antimicrobial agents with activity against enterococci.

The rate of resistance of *enterococci* to Ciprofloxacin is higher than what was found in an earlier study in the same population in 2002⁹. This increase is possibly due to increasing use of flouroquinolones¹⁰ due to convenience of administration and spectrum of action. The rate of resistance to ciprofloxacin is higher in *E. faecalis* (65.9%) which is a more common cause of clinical infections than *E. faecium* (59.5%). This finding is worrisome and points to the need for antibiotic stewardship and policy to control the use of these agents. Even though acquired resistance of enterococci to flouroquinolones has been described¹, studies have shown that newer flouroquinolones like clinafloxacin, sparfloxacin and levofloxacin show greater in vitro activity than ciprofloxacin against enterococci.¹¹

The rates of resistance to ampicillin also shows an increase from 2002⁹. Ampicillin resistant was found to be significantly higher in *E. faecium* compared to *E. faecalis*. This is not unexpected, as *E. faecium* is traditionally known to be more resistant, though less virulent, than *E. faecalis*. This trend in virulence of enterococcal

species, however, is changing with the emergence of vancomycin-resistant enterococci most of which are *E. faecium* and causing higher rates of clinical infections than non-VRE *E. faecium*. This finding also has implication for empirical treatment of enterococcal infections. Distinction between *E. faecalis* and *E. faecium* will determine the usefulness of Ampicillin (or ampicillin combinations) in treating these infections. Hence the need for distinguishing between these species in clinical laboratory reports.

High level gentamicin resistance occurred in 38% of isolates studied. This is significantly higher than 11% of clinical isolates reported in 2002⁹. Nosocomial transmission of high-level gentamicin resistant strains of enterococcus has been demonstrated. Zervos *et al.* demonstrated the patient-to-patient transmission and inter-hospital spread of a high-level gentamicin-resistant *E. faecalis* strain¹³⁻¹⁴. Transient carriage of organisms on the hands of healthcare personnel was the suggested mode of transmission. Among the factors known to be associated with infection with high-level gentamicin resistant *E. faecalis* are prior antimicrobial therapy, perioperative antibiotic prophylaxis, pre-surgical procedures, and longer hospitalizations¹²⁻¹⁴. Proper hand hygiene in hospitals will be required to prevent potential nosocomial transmission of these organisms.

This surveillance study found one isolate with vancomycin resistance. Even though this prevalence is low, it lends credence to other reports of vancomycin resistant enterococci in Nigeria^{5,6}. This draws attention to the need for screening of enterococcal isolates for vancomycin resistance in order to rapidly institute control measures when they occur.

The significance of these findings, however, is limited by the fact that the isolates used in this study were from stool samples collected for surveillance rather than from clinical specimen. Instituting more control on the use of flouroquinolones and other classes of antibiotics in healthcare settings and in the community is required to ensure that the efficacy of currently available antimicrobial agents is preserved.

Conflict of Interest: None reported

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