# EPH - International Journal Of Medical And Health Science

ISSN (Online): 2456-6063 Volume 04 Issue 04 December 2018

#### DOI: https://doi.org/10.53555/eijmhs.v5i2.68

# VANCOMYCIN RESISTANT *ENTEROCOCCUS FAECALIS* AMONG APPARENTLY HEALTHY POPULATION IN KHARTOUM LOCALITY 2017.

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### Abstract

**Background:** Enterococcus faecalis belong to the normal bacterial flora of the gastrointestinal tract of human. Vancomycin resistant Enterococcus faecalis is due to the synthesis of modified cell wall precursors that show a decreased affinity for glycopeptides. The aim of this study was to detect the Vancomycin Resistant Enterococcus faecalis among apparently healthy Population in Khartoum locality, Sudan.

*Materials and Methods:* Three hundred and eighty five faecal specimens were collected from apparently healthy population, isolated and identified of *E*. faecalis by using routine methods. The susceptibility pattern of the *E*. faecalis to Vancomycin antibiotic was determined by using Kerby's Pauer disk diffusion method.

**Results:** Out of 385 studied specimens, 87(22.6%) of E. faecalis was isolated. The isolated strains of E. faecalis showed highly resistance to Vancomycin (14.9%).

Conclusions: The present of Enterococcus faecalis among apparently healthy and have high rate of antibiotic resistant

Keywords:- Enterococcus faecalis, Vancomycin.

# **INTRODUCTION:**

Vancomycin-resistant enterococci (VRE) have emerged as serious nosocomial pathogens, accounting for approximately 8% of enterococci infections in the United States (Fraser *et al.*, 2016).

Enterococci are part of the normal intestinal flora of humans. Now a days, enterococci have become one of the most common nosocomial pathogens they can cause significant problems in healthcare facilities in susceptible populations, with a high mortality rate of up to 61% (Fisher *et al.*, 2009; Landman *et al.*, 1996).

*Enterococcus faecalis* (*E. faecalis*) was classified amongst the Lancefield group D streptococci in the 1980s. The genus Enterococcus includes more than 17 species, *E. faecalis* is the commonest species, which represent about (80–90%) of clinical isolates. It have ability to grow in 6.5% NaCl and survive heating at 60°C (Fisher *et al.*, 2009; Landman *et al.*, 1996; Török *et al.*, 2017).

*E.faecalis* acquire resistance to many antimicrobials included Vancomycin, become raise common in the hospital setting, according to National Nosocomial Infections Surveillance (NNIS) in 2003 more than 28% enterococci isolates were Vancomycin-resistant. In addition several genes isolated from resistant enterococci (agg, gelE, ace, cylLLS, esp, cpd, fsrB) encode virulence factors such as the production of gelatinase and hemolysin (Landman *et al.*, 1996).

*E. faecalis* is able to cause infections to human specially in children and elderly include urinary tract infection, endocarditis, bacteremia, catheter-related infections, surgical wound infections, and intra-abdominal and pelvic infections, Meningitis, pleural space infections and neonatal sepsis (Török *et al.*, 2017).

*E. faecalis* have both mechanism an intrinsic and acquired, which making them important nosocomial pathogens. Acquired resistance occur on transfer the genes through the exchange of transposon present on the chromosome and carried on a plasmid such as Glycopeptide (Landman *et al.*, 1996). Glycopeptide-Resistant *E. faecalis* are resistant to bactericidal antibiotic against gram-positive bacteria (Vancomycin and Teicoplanin). It is widely distributed but has poor CSF penetration in the absence of meningeal inflammation. It is excrete unchanged in the kidneys and dose reduction is required in renal impairment, Vancomycin resistant *E. faecalis (VREF)* was first ever reported in 1988 in the UK and in USA in 1989 and normally found on human intestinal tract without cause disease unless in some condition. It's include two component system where the cell wall composition is alter from the peptidoglycan precursor D-Ala-D-Ala (Vancomycin, susceptible) to D-Ala-D-lactate (D-Lac) (replacement of D-alanine in construction of peptide and glycan in the cell wall) that less affinity for Vancomycin, while D-Ala-D-Ser has a seven fold decrease in affinity for Vancomycin, thus removing the susceptible target. The genes involved in this two-component system are vanS/vanR, The vanS sensor kinase is activate in response to Vancomycin, resulting in the activation of D-Lac or D-Ser peptidoglycan precursor and the repression of D-Ala-D-Ala (Landman *et al.*, 1996; Török *et al.*, 2017).

VREF have threatened human health, as Vancomycin is a common therapeutic used for severe enterococci infections. Nevertheless, clinically relevant infections are rarely report. However, even vancomycin-susceptible enterococci may be resistant to a wide range of antimicrobials, leaving few viable treatment options in some cases (Weese, 2015; Kwon *et al.*, 2012).

In Nigeria a study on 73 Enterococcus were phenotypically identified and 65 of the isolates were 29 (44.6%) *E. faecalis*, isolated strains was resistant to vancomycin (13.8%). Remarkable multiple antibiotic resistances to the classes of antibiotic tested were observed (Adesida *et al.*, 2017)

In Europe, a study on vancomycin resistant *E. faecalis*, three countries (Ireland, Luxembourg and Greece) reported that the rate of resistant above 25% and other five countries reported resistant between 10% and 25%, while the majority of countries (18 of 26) reported resistant proportions below 10%. Several countries reported even below 1% (Bulgaria, Estonia, Finland, France, Norway, Romania and Sweden) (European Central for Disease and Control, 2009).

In 2012 in Iran, a study on 91 *E. faecalis*. Among all isolates, 15 (16%) were identified as VR *E. faecalis* (Pourakbari *et al.*, 2012).

In Kuwait, a study on 415 enterococci isolates was obtained from different clinical samples involve stool. About (85.3%) from isolated were *E. faecalis*, it was showing resistant to Vancomycin (1.9%). There was no evidence of clonal spread of the vancomycin-resistant isolates (Udo *et al.*, 2003).

In Portugal, a study on 247 faecal enterococci isolates from 99 healthy Portuguese individuals revealed the presence of *E. faecalis* resistant to vancomycin (5) (Novais *et al.*, 2006).

In India, a study on a total 180 enterococci isolates were studied. *E. faecalis* was 108 (60%) as commonest. The resistance rates to *E. faecalis* isolated to Vancomycin 5 (4.6%) (Gangurde *et al.*, 2014).

In Greek, a study on 263 fecal samples was collected from 97 infants after delivery. About 54.6% of isolated was *E. faecalis*. The isolates were examined for their resistance and showed resistant to vancomycin (11.2%) (Kirtzalidou *et al.*, 2012).

In Germany, a study on antibiotic resistance of 730 enterococci isolates. *E. faecalis* was the leading enterococci species 88.8%, resistance rates to Vancomycin (0.5%) (Reinert *et al.*, 1999).

## Materials and methods:

A descriptive cross-sectional community based study; three hundred and eighty five faecal specimens were collected from apparently healthy participants in Khartoum city and inoculated on Bile Esculin agar (Himedia, India) then incubated at 37 °C aerobically for 24-48 hours.

Colonial morphology, Gram's staining and biochemical tests (Catalase production, growth in nutrient broth containing 6.5% NaCl, Litmus Milk test and Carbohydrate Fermentation (Mannitol and Arabinose (BDH Laboratory, England)) for identified *E. faecalis* isolates (Leboffe *et al.*, 2011; Cheesbrough, 2006).

Antimicrobial susceptibility test:

Kirby-Bauer disc diffusion method was used to detect susceptibility of microorganism against antibiotic as Vancomycin (30µg), (Himedia, India) were tested. The diameter of inhibition zone around discs were measured and compared with zone of inhibition of *E. faecalis* (ATCC 29212) control strain) (Hudzicki, 2013).

## Data analysis:

Study data was processed by using Microsoft Excel version 2013 and analysed frequency tables were did by using Statistical Package for Social Sciences (SPSS.) version 25 software computer package.

#### **Ethical consideration:**

Ethical approval were obtained from Sudan International University management, verbal consents from each participant and Khartoum state ministry of health research department.

#### **Results:**

Out of the 385 studied faecal specimens, 87 *E. faecalis* strains were isolated (22.6%). The distribution of isolates between male and female was 51 (58.6%) and 36 (41.2%) respectively. Distribution of isolates within different occupation was 26 (29.9%), 6 (6.9%), 15(17.2%), 13(15%), 7(8%), 9 (10.3%), 3 (3.4%) and 8 (9.2%) among University student, Chief, Employee, Mechanical Engineer automotive, Idlest, Housewife, Plumber, and woman tea seller respectively.

The isolated strains of *E. faecalis* showed highly resistance to Vancomycin (14.9%). On the other hand sensitive strain were classified into sensitive and intermediate sensitive according to diameter of inhibition zone, sensitivity to Vancomycin were detected in (55.2%) and intermediate sensitivity to Vancomycin about (29.9). (Table (1).

Ten (76.9%) out of 13 vancomycin resistant *E. faecalis*, 10 were isolated from male while three (23.1%) were isolated from female.

Table (1) Antimicrobial susceptibility of Enterococcus faecalis Isolates to antimicrobial agents

Sensitive		Inte	ermediate	Resistant
Vancomycin	48 (55.2%)	26 (29.9%)	13 (14.9%) <b>D</b>	iscussion:

*E. faecalis* has emerged in recent years, as well as acquired resistance to many antimicrobials such as vancomycin, resulting in multidrug-resistant strains, which become increasingly common in the hospital setting (Fraser *et al.*, 2016; Fisher *et al.*, 2009).

The present study investigated the prevalence and antibacterial resistance patterns of *E. faecalis* isolate from faecal sample of apparently Healthy Population in Khartoum – Sudan.

Out of 87 isolates, 14.9 % was resistance to Vancomycin this was slightly similar to Adesida1 *et al* (Nigeria 2017) 10.4%, Kirtzalidou *et al* (Greek, 2012) 11.2% and surveillances report on five countries in Europe (European Central for Disease and Control, 2009) 10 - 25%.

It was lower than surveillances report on three countries in Europe (European Central for Disease and Control, 2009) above 25% and Pourakbari et al (Iran, 2012) 15%. While higher than Udo et al (Kuwait, 2002) 1.9%, Novais et al (Portugal, 2001) 5%, Gangurde et al (India, 2013) 4.6%, surveillances report on majority of countries (18 of 26) (European Central for Disease and Control, 2009) below 10 even below 1% and Reinert et al (Germany 1997) 0.5%.

The variation in isolation rate and resistance of E. faecalis may be due to differences in the locality of study area, population (immunity, diet and antibiotic in-take) and protocol of antimicrobial uses

#### Conclusion:

The present of Enterococcus faecalis among apparently healthy and have high rate of antibiotic resistant.

#### **Recommendations:**

The high rate of vancomycin resistant *Enterococcus faecalis* this might increase the chance to transfer the resistant gent to another bacteria, so the institute should be a regular implemented surveillance to detect emerging resistance.

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