Evaluation of chromogenic VRE medium versus Conventional Vancomycin E test in Detection of Vancomycin Resistant Enterococci

Thesis

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Abbreviations

AFLP	:	Amplified Fragment Length of polymorphism
AS	:	Aggregation Substance
BE	:	Bile Esculin
BEAV	:	Bile Esculin Azide Vancomycin
CFU	:	Colony forming unit
CAN	:	Columbia colistin nalidixic acid agar
CLSI	:	Clinical and laboratory standard institute
DNA	:	Deoxyribonucleic acid
EB	:	Enterococcosel broth
ECM	:	Extra Cellular Matrix
ESP	:	Enterococcal Surface Protein
HICPAC	:	Hospital Infection Control Practices Advisory Committee
MIC	:	Minimal inhibitory concenteration
MLST	:	Multilocus Sequence Typing
NAG	:	N-acetylglucosamine
MRSA	:	Methicillin resistant *staphylococcus aureus*
MRSE	:	Methicillin resistant *Staphylococcus epidermidis*
NAM	:	N- acetyl muramic
NaCl	:	Sodium chloride
NCCLS	:	National Committee for Clinical Laboratory Standards
NNIS	:	National Nosocomial Infections Surveillance
ORF	:	Open reading frames

Abbreviations (Cont.)

PAI	:	Pathogenicity islands
PMN	:	Poly morphonuclear cells
PBP	:	Penicillin binding proteins
PCR	:	polymerase chain reaction
PFGE	:	Pulsed Field Gel Electrophoresis
ROS	:	Reactive Oxygen Species
rRNA	:	Ribosomal ribonucleic acid
VA	:	Vancomycin
VRE	:	Vancomycin Resistant Enterococci
VRSA	:	Vancomycin resistant *staphylococcus aureus*
VISA	:	Vancomycin intermediate resistant *Staphylcoccus aureus*
VSE	:	Vancomycin sensitive enterococci
WCP	:	Whole cell protein

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Abstract

Enterococci have become resistant to a wide range of antibiotics which include glycopeptides like vancomycin. The rapid increase of vancomycin resistance compromises physicians' ability to treat infections caused by these strains because the therapeutic options for VRE infections are very limited. Our aim in this study was to evaluate the efficacy of chromogenic VRE medium in detection and identification of vancomycin resistant enterococci.

The present study was conducted in Al Abasia Fever Hospital, El Sayed Galal Hospital, Tanta University Hospital and Kafr El Sheikh General Hospital. Sixty enterococcal isolates were collected (thirty vancomycin susceptible, eight ancomycin intermediate resistant and twenty two vancomycin resistant isolates). Identification of these isolates to genus level was confirmed. Antibiotic susceptibility pattern for enterococcal isolates was done using disc diffusion method, Chromogenic medium and The MIC of vancomycin for vancomycin resistant and intermediate resistant isolates was determined by E test.

Identification of vancomycin resistant and intermediate resistant isolates to species level was done by HiStrep rapid identification kit and the VITEK-2 COMPACT system to confirm species identification of these isolates obtained by chromogenic media.

The most common isolated vancomycin resistant and intermediate resistant species was *E. faecium* (53.1%) followed by *E. faecalis* (40.6%). The highest rate of isolation of VRE was from urine (50.0%). VRE isolates were mostly recovered from ICUs (45.5), 41.7%, of the collected isolates were vancomycin resistant by disc diffusion method. Regarding E-test, out of fourty two vancomycin resistant and intermediate resistant by disc diffusion method 52.4% were resistant and 19% were intermediate resistant. HiChrome VRE had 100% sensitivity and 83.3% specificity in identifying vancomycin resistant enterococcal isolates, it have 100% sensitivity and specificity in identifying *VRE. faecalis* and *VRE. faecium*.

Conclusion: We can depend on chromogenic media in both detection and identification of vancomycin resistant enterococci.

Key words: Enterococci, Multidrug resistant enterococci, VRE, Chromogenic media, Infection control of VRE.

INTRODUCTION

Enterococcus species are members of the normal intestinal flora and are the most common aerobic Gram-positive cocci found in the large bowel of humans. Enterococci had become an important nosocomial pathogens that were grouped as the third most common cause of bacteremia in the United States ***(Centers for Disease Control and Prevention, 1999).***

Of the clinically important entero-cocci is *enterococcus faecium* (*E.faecium*) which is more likely to acquire resistance to glycopeptides also is linked more frequently to serious disease (***Moellering, 2005***).

Vancomycin-resistant enterococci (VRE) have emerged as important pathogens in many health care facilities. VRE may lead to extra intestinal infections, such as bacteremia and peritonitis (***Shigei et al., 2002***).

One of the important virulence factors in enterococci is their resistance to several antimicrobial agents and this resistance can be intrinsic (low level to penicillin, cephalosporins and aminoglycoside) as well as acquired resistance to glycopeptides an high level resistance to aminoglycosides ***(Mathur et al., 2003).***

Prevalence rates of vancomycin-resistant enterococci (VRE) associated with serious clinical infections have increased worldwide over the past fifteen years ***(Deshpande et al., 2007*)**. An obstacle to control of the spread of VRE is the large, unrecognized cohort of patients with gastro intestinal VRE colonization ***(Tacconelli and Cataldo, 2008*)**. Therefore,

prompt accurate identification of patients with VRE is imperative. Thus to prevent further spread; active control measures are increasingly being implemented in hospitals. A cornerstone of those control measures is the detection of non infected but gut-colonized patients that might serve as a source of the spread of VRE (***Vijaya et al., 2014).***

The clinical impact of infection by VRE has been examined in several studies, with the most notable consequences being increases in mortality, length of hospital stay, and cost of hospitalization. While a clear link between colonization with glycopeptide-resistant enterococcus and increased mortality has not been clearly established (***Moellering, 2005***). Antimicrobial therapy does promote selection and proliferation of VRE in the hospital environment (***Moellering, 1992***). Patients who become colonized with VRE can become asymptomatic carriers for months to years (***Moellering, 2005***).

Detection of VRE colonization relied on culture techniques using selective/differential media **(*Teixeira and facklam, 2003*).** Bile esculin azide agar supplemented with 6gm of vancomycin/ml (BEAV) was the most effective medium identified for screening for VRE **(*Moellering, 1992).*** Furthermore, the use of selective culture techniques can be time consuming usually require 24 to 48 h to preliminarily identify colonies, with additional time for confirmatory identification and susceptibility testing. Molecular VRE screening methods decrease the time to identification but are costly. Culture remains the screening method of choice for VRE stool screening **(*Stamper et al.*, 2007*).***

Various chromogenic VRE agars appear promising for use in VRE stool screening ***(Cuzon et al., 2008).*** Chromogenic VRE media can reduce turn around time to results through early visual colony identification of VRE direct from clinical samples ***(Hajia et al., 2012).***

Chromogenic media are increasingly used as versatile tools in early differentiation and identification of VRE from clinical samples ***(Hajia et al., 2012).*** Another similar study was previously done to evaluate chromogenic VRE medium in detecting VRE in comparison with E- test **(*Vijaya et al., 2014*).**