Profile of Bacterias Isolated in Blood Culture by Automated System at a Tertiary Care Hospital in Northern India

Sikander Chirag¹, Suman Kumar Kotwal², Shalija Kotwal³, Bashir Famda⁴, Sahrish Chirag⁵

IJMHS (Jul-Dec 2017) 04 (2): 79-82 / ©Red Flower Publication Pvt. Ltd.

Abstract

Sepsis is one of the most common causes of death worldwide both in hospitalized and non hospitalized patients. Vitek 2 is an automated system used for identification of bacterial isolates. These automated systems in bacteriology are very important for rapid and accurate detection of bacterias for effective diagnosis and treatment. Vitek 2 identified 18 genera of bacterias, 40 up to species level and 4 up to subspecies level. A total of 299 blood culture were tested, most common organism were gram negative bacteria (fermenters) followed by nonfermenters, gram-positive cocci and some of the bacterias which were isolated were uncommon and not detected by routine culture techniques. This retrospective study was carried out in order to study the incidence of bacterias isolated from blood cultures by Vitek 2.

Keywords: Vitek 2; Sepsis; Bacterias.

Introduction

Sepsis is a life threatening condition that arises

Author's Affiliation: ¹Lecturer, Department of Microbiology ²Consultant, Department of Medicine ³Senior Resident, Department of Pathology, Government Medical College, Jammu, Jammu and Kashmir 180001, India. ⁴Consultant, Department of Microbiology, Sher-I-Kashmir Institute of Medical Sciences, Srinagar, Jammu & Kashmir-190011, India. ⁵Department of Animal Husbandry, Jammu, Jammu and Kashmir 180001.

Corresponding Author: Suman Kumar Kotwal, Consultant, Department of Medicine, Government Medical College, Jammu, Jammu and Kashmir 180001, India.

E-mail: sumankk1230@rediffmail.com

Received on 30 April 2017 Accepted on 09 May 2017 when the body response to infection causes injury to its own tissues and organs [1]. Common signs and symptoms include fever, increased breathing rate and confusion [2]. There may also be symptoms related to specific infection such as cough, pneumonia etc. Severe sepsis is sepsis associated with poor organ function. Sepsis is caused by an immune response triggered by infection [3, 4].

Blood cultures are very necessary for diagnosis of sepsis and for prompt administration of antibiotics. Patients with positive blood cultures are 10 times more likely to die during hospitalization than those with negative blood cultures [5]. In the past microbiology laboratories rely on conventional blood culture procedures for identification of bacterias and antimicrobial susceptibility, but accuracy of these customary procedures was far inferior to automated detection systems which can distinguish even up to subspecies level.

This automated system (Vitek 2) automatically performs all steps required for identification and antimicrobial susceptibility testing after primary inoculums have been prepared and standardized [6]. Vitek 2 system allows kinetic analysis by reading each test every 20 minutes. The optical system combines multichannel flourimeter and photometer readings to record fluorescence, turbidity and calorimetric signals [7]. Use of this automated technique allows quicker, faster and accurate detection of bacterias.

Results

A total of 299 positive blood culture specimens were tested and identified by Vitek 2 system. Of these positive specimens 20 genera, 39 species were also registered. In our study we have also seen uncommon bacterias which are often difficult to detect by

conventional culture. These were nonfermenting gram-negative bacterias like alcaligenes faecalis, sphingomonas paucimobilis, methylobacterium spp, chyrsobacterium indologenes, burkholderia cepacia and gram positive cocci like Leuconostoc mesenteroides and aerococcus viridians. (Table 1 and 2).

Materials and Methods

This study was retrospectively conducted at tertiary care hospital in northern India. During the study received blood culture bottles were loaded into the automated system BacT/ALERT3D(biomerieux),

Table 1: Profile of gram negative bacterias on vitek 2 system in cases of sepsis

Gram Negative Bacteria	
Subtype	No. of Bacteria
Enterobacteriacae	
E.coli	65
Klebsiella pneumonia ssp pneumonia	57
Klebsiella sp	1
Enterobacter cloacae complex	4
Enterobacter cloacae ssp dissolvens	47
E.hartmanii	1
Citrobacter freundi	3
Proteus mirablis	3
Proteus vulgaris	2
Salmonella enteric ssp enteric	2
Salmonella serotype typhi	5
Salmonella ser paratyphi	1
Serratia marcescens	1
Morganella morganii ssp morgani	2
Oxidase positive fermenter-Alcaligenes faecalis	1

Gram Negative Bacteria	
Subtype	No. of Bacteria
Non Fermenters	
Psedomonas aeruginosa	5
Psedomonas fluorescens	2
Psedomonas putida	4
Pseudomonas stutzeri	1
Burkholderia cepacia	1
Sphingomonas paucimobilis	2
Alcaligenes fecalis	1
Acinetobacter baumanii	34
Acinetobacter baumanii complex	6
Acinetobacter iwoffii	2
Acinetobacter haemolyticus	2
Acinetobacter junii	1
Methylobacterium spp	1
Chryseobacterium indologenes	1

Table 2: Profile of gram positive bacterias on Vitek 2 system in cases of sepsis.

Gram Positive COCCI	
	No. of bacteria
Staphylococcus aureus	18
Staphylococcus lugdunensis	1
Aerococcus viridians	1
Staphylococcus sciuri	1
Staphylococcus epidermidis	5
Staphylococcus haemolyticus	6
Staph hominis ssp hominis	3
Micrococcus luteus	2
Streptococcus pyogenes	3
Strep mutans	2

positive blood culture bottles were subcultured on three types of media: blood agar, macconkey and chocolate agar. Blood agar and macconkey plates were incubated at 37°C under aerobic conditions while chocolate agar incubated under microaerophilic condition by using anaerobic jar with gaspak at 37°C. The plates were examined for growth after 24-48 hours of incubation and the result of bacterial identification was recorded. The identification with vitek 2 includes ID-GN card for gram negative bacilli and ID-GP card for gram positive bacteria.

Discussion

Sepsis is a global health problem that carries a high risk of death. UNICEF announced that more than 40% of under five deaths occur in neonatal period resulting in 3.1 million newborn deaths each year due to sepsis [8].

The type of organism causing severe sepsis is an important determinant of outcome [9]. An epidemiological study on sepsis done in USA proved that bacteria are the most common causative microorganisms in sepsis [10]. Rapid and reliable identification of these organisms is essential for accurate diagnosis and prompt effective treatment of these infections [11,12]. Traditional methods of bacterial identification which rely on phenotypic identification (differential staining, culturing on specific media and some biochemical methods) is not sufficient to reach the final identification of some uncommon genera and most of species because some strains require more specific culture media and biochemical tests not routinely available in all laboratories. Use of vitek 2 helped in detection of some uncommon bacterias whose identification is crucial in patient care. This automated system (Vitek 2) can be operated even by lab technician and the main work of microbiologist is to explain the significance of reported bacteria to the concerned physician or pediatrician. Many peer reviewed publications demonstrated that automated vitek 2 technology and vitek 2 ID cards provide reliable and accurate results for clinically important gram positive cocci and gram negative bacilli [13-15].

In this study vitek 2 identified 21 generas and 39 species. In this study vitek 2 shows advantage of detection of some uncommon bacterias which included Burkhoderia cepacia, Alcaligenes fecalis, chrysobacterium indologenes, Methylobacterium species, sphingomonas paucimobilis, Aerococcus viridians, which have never been diagnosed in our centre by conventional methods. It has been

mentioned that all known pathogenic and non pathogenic (opportunistic) species have been isolated from blood in particular cases [16]. Improper use of antibiotics has led to emergence of opportunistic bacterias, which are threat in immunocompromised population. Vitek 2 also led to identification of coagulase negative staphylococci (CONS) which are again difficult to detect in conventional laboratory because of lack of all biochemicals required. These coagulase negative staphylococci play a role in nososcomial blood stream infections [17,18]. Coagulase negative staphylococci which were once considered commensals organisms of human skin and mucous membranes are now opportunistic pathogens in ICU [19,20].

In our study more gram negative organisms were isolated than gram positive in blood cultures, similar findings were shown in studies conducted in Iran and Saudi Arabia [21]. In our study CONS, E.coli, Klebsiella pneumonia, pseudomonas spp, Acinetobacter spp to bacter spp were most commonly isolated in blood stream infections. More or less similar observations have been made in cases of bacteremia in different countries [22].

References

- Singer M, Deutschman CS, Seymour CW, Shankar-Hari M, Annane D, Bauer M, Bellomo R, Bernard GR, Chiche JD, Coopersmith CM, Hotchkiss RS, Levy MM, Marshall JC, Martin GS, Opal SM, Rubenfeld GD, van der Poll T, Vincent JL, Angus DC. "The Third International Consensus Definitions for Sepsis and Septic Shock (Sepsis-3)". JAMA. 2016;315: 801–810.
- Centers for Disease Control and Prevention (CDC). May 22, 2014. Retrieved 28 November 2014.
- Jui, Jonathan. "Ch. 146: Septic Shock". In Tintinalli, Judith E.; Stapczynski, J. Stephan; Ma, O. John; Cline, David M.; et al. Tintinalli's Emergency Medicine: A Comprehensive Study Guide (7th ed.). New York: McGraw-Hill. 2011.p.1003-14.
- Deutschman, CS; Tracey. "Sepsis: Current dogma and new perspectives". Immunity. 2014;40(4):46375.
- 5. Bryan, C.S. Clinical implications of positive blood cultures. Clin Microbiol Rev., 1989;2(4):329–353.
- 6. Funke, G., Monnet, C. deBernardis, A. von Graevenitz, and J. Freney. Evaluation of the VITEK 2 system for rapid identification of medically relevant gram-negative rods. Clin. Microbiol. 1998;36:1948-1952.
- Marco Ligozzi, Cinzia Bernini, Maria Grazia Bonora, Maria de Fatima, Jessica Zuliani, and Roberta Fontana. Evaluation of the VITEK 2 System for Identification and Antimicrobial Susceptibility

- Testing of Medically Relevant Gram-Positive Cocci. Clin Microbiol. 2002;40(5):1681–1686.
- 8. UNICEF, WHO, The World Bank, and The United Nations. 2011. Levels and Trends in Child Mortality, UNICEF, New York, NY, USA.
- Vincent, JL., Rello, J., Marshall, J., Silva, E., Anzueto, A., Martin, CD., Moreno, R., Lipman, J., Gomersall, C., Sakr, Y. EPIC II Group of Investigators International study of the prevalence and outcomes of infection in intensive care units. AMA. 2009;302: 2323–9.
- Martin GS, Mannino DM, Eaton S, Moss M. The epidemiology of sepsis in the United States from 1979 through 2000. N Engl J Med., 2003;348:1546-1554.
- 11. Paul, M., Shani, V., Muchtar, E., Kariv, G., Robenshtok, E., Leibovic, L. Systematic review and meta-analysis of the efficacy of appropriate empiric antibiotic therapy for sepsis. Antimicrob Agents Chemother. 2010;54:4851-4863.
- 12. Barenfanger, J., Short, M. A. and Groesch, A. A. Improved antimicrobial interventions have benefits. Clin. Microbiol. 2001;39:2823-2828
- 13. Funke, G., and Funke-Kissling, P. Performance of the New VITEK 2 GP Card for Identification of Medically Relevant Gram-Positive Cocci in a Routine Clinical Laboratory. Clin. Microbiol. 2005;43(1):84-88.
- 14. Marco, L, Cinzia, B, Maria, G. B., Jessica, Z. and Roberta, F. Evaluation of the VITEK2 System for Identification and Antimicrobial Susceptibility Testing of Medically Relevant Gram-Positive Cocci. Clin. Microbiol. 2002;40(5):1681-1686.

- 15. Higgins, C. Laboratory diagnosis of sepsis: blood culture and beyond. Lancet. 2010;375:224–230.
- Kloos WE, Bannerman TL. Update on clinical significance of coagulase-negative staphylococci. Clin. Microbiol. Rev. 1994;7:117–140.
- 17. Lang S, Livesley MA, Lambert PA, Elliott J, Elliott TS. The genomic diversity of coagulase- negative staphylococci associated with nosocomial infections. Hosp. Infect. 1999;43:187–193.
- 18. Grice, EA., Segre, JA. The skin microbiome. Nat. Rev. Microbiol. 2011;9:244–253.
- Petti, CA., Simmon KE, Miro JM, Hoen B, Marco F, Chu VH, Athan E, Bukovski S, Bouza E, Bradley S, Fowler VG, Giannitsioti E, Gordon D, Reinbott P. 2008. Genotypic diversity of coagulase-negative staphylococci causing endocarditis: a global perspective. Clin. Microbiol. 2008;46:1780-1784.
- Krediet TG, Mascini EM, vanRooij E, Vlooswijk J, Paauw A, Gerards LJ, Fleer A. Molecular epidemiology of coagulase-negative staphylococci causing sepsis in a neonatal intensive care unit over an 11-year period. Clin. Microbiol. 2004;42:992–995.
- 21. Rahbar M, Gra-Agaji R, Hashemi S. Nosocomial blood stream infections in Inam Khomeini Hospital, Urmia, Islamic Khomeini Hospital, Urmia, Islamic Repulic of Iran, 1999-2001 East Mediterr Health J 2005 May;11(3):484.
- 22. Emori TG, Gaynes RP. An overview of nosocomial infections, including the role of the microbiology laboratory. Cin Microbiol Rev. 1993;4:428-442.