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Evaluation of GeneXpert vanA/vanB assay for the detection of vancomycin-resistant enterococci in patients newly admitted to intensive care units

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Aim: The automated Cepheid GeneXpert system provides rapid PCR results and can be used for the identification of VRE. We aimed to evaluate the use of the Cepheid Xpert vanA/vanB real-time PCR assay for the detection of VRE from rectal swabs of patients newly admitted to intensive care units in a hospital setting.

Materials and methods: Rectal swab samples of patients newly admitted to 6 intensive care units from March 2011 to February 2012 were obtained. The specimens were analyzed by the GeneXpert system. The results were reported for both vanA and vanB as negative or positive.

Results: Comparing the number of inpatients, most of the samples were delivered from the neurosurgery (48.3%), pediatrics (33.3%), and neonatology (20.6%) intensive care units. The positive samples according to GeneXpert vanA/vanB method were 33 (7.3%) among 454 rectal samples. Of these positive samples 22 (4.9%) were vanA, 10 (2.2%) were vanB, and 1 sample (0.2%) was vanA and vanB-harboring, by PCR.

Conclusion: As a rapid, easy to use, and labor-saving method GeneXpert vanA/vanB can detect VRE-positive patients, particularly in risk groups, as soon as they are admitted to hospital so that infection control policies can be applied immediately.

Key words: Vancomycin-resistant enterococci, GeneXpert vanA/vanB assay, intensive care unit

1. Introduction

Enterococci are one of the leading causes of nosocomial infections. In recent years enterococci have become increasingly resistant to a wide range of antimicrobial agents. In addition, vancomycin-resistant enterococci (VRE) have become resistant to glycopeptide antibiotics. Glycopeptide-resistant enterococci have become a main hazard to hospitalized patients. Similar to methicillin-resistant *Staphylococcus aureus*, VRE can give rise to significant nosocomial epidemics and can raise morbidity, mortality, and costs related to admission to the hospital (1,2).

Patients monitored in a medical intensive care unit (ICU) have a high risk of VRE colonization/infection, and active VRE surveillance of high-risk group patients is crucial for early detection and implementation of precautions to impede the development of infection and the spread of VRE (3,4).

Rapid and accurate microbiologic identification of VRE is essential for the management of both colonized and infected patients in order to select adequate treatment and to prevent the spread of VRE by implementing proper barrier precautions. VRE are classically screened by culture-based processes, considered the "gold standard," which are time consuming (48–72 h), and other phenotypic investigations are needed. Nucleic acid amplification tests can be used for the detection of VRE, but complicated extraction and detection steps are required (5–7). Moreover, a culture step from a selective enrichment broth or solid media may be needed for some methods (8,9). Recently, automated real-time PCR in vitro test assays for the rapid detection of vancomycin resistance, directly from perianal or rectal swabs, came into use (10,11). The GeneXpert system (Cepheid, Sunnyvale, CA, USA) merges automated nucleic acid sample preparation, amplification, and real-time detection of enterococcus DNA in a disposable, macro/microfluidic cartridge using the GeneXpert Dx system instrument and generally provides results in less than 1 h (12).

The aim of this study was to evaluate the use of the Cepheid Xpert vanA/vanB real-time PCR assay for the detection of VRE from rectal swabs of patients newly admitted to ICUs in a hospital setting.

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2. Materials and methods

Patients newly admitted to ICUs from March 2011 to February 2012 were included in the study. Routine surveillance by culture method was also carried out on all the patients. The 6 different ICUs involved in the study were anesthesiology and reanimation, neurology, internal medicine, neurosurgery, pediatrics, and neonatology. Rectal swab samples were collected from each patient using Amies transport medium (Meus, Italy) and transported to the laboratory. The samples were analyzed by automated multiplex real-time PCR assay (GeneXpert vanA/vanB, Cepheid, Sunnyvale, CA, USA) according to the recommendations of the manufacturer. Briefly, the swab sample obtained from the patient was added to the elution buffer and vortexed for 1 min. The buffer with the swab was transferred to a single-use disposable cartridge containing integrated chambers and reagents. Then the cartridge was placed in the GeneXpert™ Dx module and run. The results were reported for both vanA and vanB as negative or positive. When only vanB was positive, the culture method was used as a confirmatory test.

3. Results

Over a period of 11 months rectal swab samples from 454 patients were obtained, and VRE colonization was evaluated. Comparing the number of inpatients, most of the samples were delivered from the neurosurgery (48.3%), pediatrics (33.3%), and neonatology (20.6%) ICUs, respectively. However, samples that showed the presence of VRE by multiplex real-time PCR were low (6.9%, 5.3%, and 2.8%, respectively). Although the number of delivered samples was low (7.3%) compared to the total number of inpatients in the anesthesiology and reanimation ICUs, VRE by PCR was detected in 36% of the samples. Furthermore, multiplex real-time PCR detected 22 (4.9%) samples that showed only vanA- harboring VRE, 10 (2.2%) with only vanB-harboring VRE, and 1 sample (0.2%) with both vanA and vanB-harboring VRE (Table). Therefore, the presence of vanB was seen in 11 samples (2.4%) in total. Five of the patients harboring vanB only were negative by the culture method, and the remaining 5 could not be evaluated by culture because they died or were referred to another hospital.

4. Discussion

The Gene Xpert vanA/vanB assay was recently described as a rapid and accurate method for detecting VRE from perianal/rectal swabs of colonized or infected patients. This fully automated process combines DNA extraction, real-time PCR amplification, and detection steps, and the results are obtained generally in less than 1 h (10,12,13). In the first report using the assay for detecting VRE the sensitivity and negative predictive value of the method were 100% compared to enriched culture, and it was indicated that this method could be used to control only positive PCR results in order to reduce laboratory labor (12). This assay has a higher sensitivity for the detection of both vanA and vanB-harboring VRE at lower bacterial loads (10–100 cfu/mL) and could also provide rapid detection of VRE carriage in patients at the time of hospital admission in conjunction

Table. Vancomycin-resistant enterococci (VRE) positive results of samples obtained from 6 different intensive care units (ICUs) by Cepheid Xpert vanA/vanB assay.

	Number of inpatients	Obtained samples n (%)	VRE positive		
			VanA positive n	VanB positive n	Total n (%)
AR-ICU	336	25 (7.4)	7	2	9 (36)
NEU-ICU	324	32 (9.9)	1	1	2 (6.3)
IM-ICU	647	56 (8.7)	3	1	4 (7.1)
NC-ICU	362	175 (48.3)	7	5	11 (6.3)
P-ICU	285	95 (33.3)	3	2	5 (5.3)
NE-ICU	344	71 (20.6)	2	-	2 (2.8)
TOTAL	**2298**	**454 (19.7)**	**23**	**11**	**33 (7.3)**

VRE: vancomycin-resistant enterococci
AR-ICU: Anesthesiology and Reanimation Intensive Care Unit
NEU-ICU: Neurology Intensive Care Unit
IM-ICU: Internal Medicine Intensive Care Unit
NC-ICU: Neurochirurgie Intensive Care Unit
P-ICU: Pediatrics Intensive Care Unit
NE-ICU: Neonatology Intensive Care Unit

with culture confirmation (14). Therefore, we chose this method to detect VRE-positive patients as soon as they were admitted to ICUs in our hospital in order to apply infection control policies immediately. Patients with a positive result for VRE are reported to infection control nurses.

In the study period we collected specimens from 454 (19.8%) patients among 2298 total ICU inpatients (Table). Most of the samples were obtained from the neurosurgery ICU; this may be because these patients stay for a short period of time after surgery and so patient turnover was high compared to other ICUs. We requested rectal swab samples from all patients who were newly admitted to the ICUs; however, some of the units sent only a small portion of all samples. In particular, the proportion of specimens obtained from the anesthesiology and reanimation, neurology, and internal medicine ICUs were lower when compared to the number of inpatients (7.3%, 9.9%, and 8.7%, respectively) (Table). Continuing routine weekly surveillance by culture method may be one of the reasons for this. The intensive care staff may have been reluctant to send extra samples, because this type of specimen collection can be offensive. Informing the intensive care staff about the importance of early detection of VRE could increase the sample sending rate.

The prevalence of VRE carriage on ICU admission has been reported between 1.4% and 25% in different studies (12,15–20). The number of positive samples by GeneXpert vanA/vanB method was 33 (7.3%) among the 454 rectal specimens sent in our study. Some of the risk factors for VRE carriage are antibiotic use and hospitalization (20–22). The positivity rate was rather high in samples from anesthesiology and reanimation ICU (36%). The patients in this unit have been given extensive antibiotics and had the most severe status and hospitalization history. Therefore, these patients' characteristics may be the reason for the high rate of VRE positivity. Conversely, the patients in neonatology ICU have a lower rate of antibiotic use and a shorter hospitalization history. These conditions may result in the low VRE positivity.

One limitation of this study was that all vanB-positive patients were not compared with culture method, due to reasons beyond our control. The Gene Xpert vanA/vanB assay has improved sensitivity compared to direct cultures; therefore, labor-intensive broth-enrichment is not required (10). However, the results of GeneXpert vanA/vanB have a very low PPV and should be confirmed by culture, especially the vanB gene (23). The possible explanations for the false-positive reactions for vanB were lack of specificity of primers/probes of the PCR assay or the presence of van genes in uncultured bacteria (12). Furthermore, when stool is present on rectal swabs, it may carry risk of vanB detection from aerobic and anaerobic bacteria of stool flora and contain PCR inhibitors (24). To avoid inhibition of amplification, samples need to be prepared carefully (13). The Gene Xpert vanA/vanB system gives an error message when stool is present on rectal swabs. When we saw this message during the process, we diluted the swab and repeated the test. Despite this, we have discrepant results between PCR and culture. Studies issuing false-positive results due to vanB suggest that follow-up culture be performed on any vanB-positive results (11,25,26). If the presence of vanB is low in the setting, culture backup may not be warranted. To assess the utility of culture confirmation for vanB, studies are ongoing (10). The presence of vanB was also low in our study group (2.4%), and so confirming vanB-positive samples by culture method may not be necessary.

There are few reports from Turkey detailing Cepheid Xpert vanA/vanB system use in the subsequent evaluation of VRE epidemics and sporadic cases (27,28). In other reports the Cepheid Xpert vanA/vanB method was evaluated for detecting VRE from rectal specimens. The results of the Cepheid Xpert vanA/vanB method have been compared to conventional culture methods in these studies (29,30). However, the administration of this system in samples of patients newly admitted to ICU had not been reported. To our knowledge this is the first report on the use of the Cepheid Xpert vanA/vanB in patients newly admitted to ICUs for detecting VRE from rectal specimens in Turkey.

When total costs including savings from a potential decrease in infections, ease of use, and laboratory labor savings are considered, the assay's relative cost-effectiveness can be estimated truly (10). Therefore, this rapid, easy to use, and labor-saving method can be used to detect VRE-positive patients in risk groups as soon as they are admitted to hospital in order to apply infection control policies immediately.

References

1. Gordts B, Van Landuyt H, Ieven M, Vandamme P, Goossens H. Vancomycin-resistant enterococci colonizing the intestinal tracts of hospitalized patients. J Clin Microbiol 1995; 33: 2842–6.
2. İncecik Ş, Saltoğlu N, Yaman A, Karayaylalı İ, Özalevli M, Gündüz M, Burgut R. The problem of antimicrobial resistance in nosocomial medical and surgical intensive care units infections in a university hospital; a two-year prospective study. Turk J Med Sci 2009; 39: 295–304.
3. Tacconelli E, Cataldo MA. Vancomycin-resistant enterococci (VRE): transmission and control. Int J Antimicrob Agents 2008; 31: 99–106.
4. Koçak Tufan Z, Arslan S, Cesur S, Bulut C, Irmak H, Kınıklı S, Ergin F, Çelik AK, Demiröz AP. Absence of vancomycin-resistant enterococci (VRE) despite the presence of risk factors: a survey of rectal carriage of VRE. Turk J Med Sci 2010; 40: 623–28.

5. Palladino S, Kay ID, Flexman JP, Boehm I, Costa AM, Lambert EJ, Christiansen KJ. Rapid detection of vanA and vanB genes directly from clinical specimens and enrichment broths by real-time multiplex PCR assay. J Clin Microbiol 2003; 41: 2483–6.

6. Paule SM, Trick WE, Tenover FC, Lankford M, Cunningham S, Stosor V, Cordell RL, Peterson LR. Comparison of PCR assay to culture for surveillance detection of vancomycin-resistant enterococci. J Clin Microbiol 2003; 41: 4805–7.

7. Satake S, Clark N, Rimland D, Nolte FS, Tenover FC. Detection of vancomycin-resistant enterococci in fecal samples by PCR. J Clin Microbiol 1997; 35: 2325–30.

8. Sahm DF, Free L, Smith C, Eveland M, Mundy LM. Rapid characterization schemes for surveillance isolates of vancomycin-resistant enterococci. J Clin Microbiol 1997; 35: 2026–30.

9. Drews SJ, Johnson G, Gharabaghi F, Roscoe M, Matlow A, Tellier R, Richardson SE. A 24-hour screening protocol for identification of vancomycin-resistant *Enterococcus faecium*. J Clin Microbiol 2006; 44: 1578–80.

10. Marner ES, Wolk DM, Carr J, Hewitt C, Dominguez LL, Kovacs T, Johnson DR, Hayden RT. Diagnostic accuracy of the Cepheid GeneXpert vanA/vanB assay ver. 1.0 to detect the vanA and vanB vancomycin resistance genes in *Enterococcus* from perianal specimens. Diagn Microbiol Infect Dis 2011; 69: 382–9.

11. Stamper PD, Cai M, Lema C, Eskey K, Carroll KC. Comparison of the BD GeneOhm VanR assay to culture for identification of vancomycin-resistant enterococci in rectal and stool specimens. J Clin Microbiol 2007; 45: 3360–5.

12. Bourdon N, Berenger R, Lepoultier R, Mouet A, Lesteven C, Borgey F, Fines-Guyon M, Leclercq R, Cattoir V. Rapid detection of vancomycin-resistant enterococci from rectal swabs by the Cepheid Xpert vanA/vanB assay. Diagn Microbiol Infect Dis 2010; 67: 291–3.

13. Wisplinghoff H, Wiegel P, Steinmetz M, Hofmann H, Arnemann J, Plum G. Evaluation of a novel rapid PCR-method to detect vancomycin-resistant enterococci (VRE) from clinical samples. 49th Intersc. Conf. Antimicrob. Agents Chemother; 12–15 September 2009; San Francisco, CA, United States. pp. D–783.

14. Gazin M, Lammens C, Goossens H, Malhotra-Kumar S, Team MWS. Evaluation of GeneOhm VanR and Xpert vanA/vanB molecular assays for the rapid detection of vancomycin-resistant enterococci. Eur J Clin Microbiol Infect Dis 2012; 31: 273–6.

15. Morgan DJ, Day HR, Furuno JP, Young A, Johnson JK, Bradham DD, Perencevich EN. Improving efficiency in active surveillance for methicillin-resistant *Staphylococcus aureus* or vancomycin-resistant *Enterococcus* at hospital admission. Infect Control Hosp Epidemiol 2010; 31: 1230–5.

16. Warren DK, Kollef MH, Seiler SM, Fridkin SK, Fraser VJ. The epidemiology of vancomycin-resistant *Enterococcus* colonization in a medical intensive care unit. Infect Control Hosp Epidemiol 2003; 24: 257–63.

17. Song JY, Cheong HJ, Jo YM, Choi WS, Noh JY, Heo JY, Kim WJ. Vancomycin-resistant *Enterococcus* colonization before admission to the intensive care unit: a clinico-epidemiologic analysis. Am J Infect Control 2009; 37: 734–40.

18. Ostrowsky BE, Venkataraman L, D'Agata EM, Gold HS, DeGirolami PC, Samore MH. Vancomycin-resistant enterococci in intensive care units: high frequency of stool carriage during a non-outbreak period. Arch Intern Med 1999; 159: 1467–72.

19. Minhas P, Perl TM, Carroll KC, Shepard JW, Shangraw KA, Fellerman D, Ziai WC. Risk factors for positive admission surveillance cultures for methicillin-resistant *Staphylococcus aureus* and vancomycin-resistant enterococci in a neurocritical care unit. Crit Care Med 2011; 39: 2322–9.

20. Yoon YK, Kim HJ, Lee WJ, Lee SE, Yang KS, Park DW, Sohn JW, Kim MJ. Clinical prediction rule for identifying patients with vancomycin-resistant enterococci (VRE) at the time of admission to the intensive care unit in a low VRE prevalence setting. J Antimicrob Chemother 2012; 67: 2963–9.

21. Donskey CJ, Chowdhry TK, Hecker MT, Hoyen CK, Hanrahan JA, Hujer AM, Hutton-Thomas RA, Whalen CC, Bonomo RA, Rice LB. Effect of antibiotic therapy on the density of vancomycin-resistant enterococci in the stool of colonized patients. N Engl J Med 2000; 343: 1925–32.

22. Muto CA, Jernigan JA, Ostrowsky BE, Richet HM, Jarvis WR, Boyce JM, Farr BM, SHEA. SHEA guideline for preventing nosocomial transmission of multidrug-resistant strains of *Staphylococcus aureus* and *Enterococcus*. Infect Control Hosp Epidemiol 2003; 24: 362–86.

23. Zabicka D, Strzelecki J, Wozniak A, Strzelecki P, Sadowy E, Kuch A, Hryniewicz W. Efficiency of the Cepheid Xpert vanA/vanB assay for screening of colonization with vancomycin-resistant enterococci during hospital outbreak. Antonie Van Leeuwenhoek 2012; 101: 671–5.

24. Ballard SA, Pertile KK, Lim M, Johnson PD, Grayson ML. Molecular characterization of vanB elements in naturally occurring gut anaerobes. Antimicrob Agents Chemother 2005; 49: 1688–94.

25. Sloan LM, Uhl JR, Vetter EA, Schleck CD, Harmsen WS, Manahan J, Thompson RL, Rosenblatt JE, Cockerill FR 3rd. Comparison of the Roche LightCycler vanA/vanB detection assay and culture for detection of vancomycin-resistant enterococci from perianal swabs. J Clin Microbiol 2004; 42: 2636–43.

26. Mehta MS, Paule SM, Hacek DM, Thomson RB, Kaul KL, Peterson LR. Optimization of a laboratory-developed test utilizing roche analyte-specific reagents for detection of *Staphylococcus aureus*, methicillin-resistant *S. aureus*, and vancomycin-resistant *Enterococcus* species. J Clin Microbiol 2008; 46: 2377–80.

27. Şen M, Gözel MG, Çelik C, Bakıcı MZ, Açıkalın Coşkun K, Özgür A, Engin A, Dökmetaş İ, Bakır M, Tutar Y et al. Sivas Cumhuriyet Üniversitesi Hastanesi'nde *Enterococcus faecium* ile oluşan bir vankomisin dirençli enterokok (VRE) epidemisi: ön rapor. 4th EKMUD Congress (Infectious Diseases & Clinical Microbiology); 9–12 May 2012; Pendik, İstanbul, Turkey. pp. 153.

28. Atalay S, Ece G, Şamlıoğlu P, Maraş G, Koşe I, Koşe S. [Evaluation of vancomycin-resistant enterococcus cases at a tertiary level hospital in İzmir, Turkey]. Mikrobiyol Bul 2012; 46: 553–9.

29. Esen Ş, Yıldız İE, Güney AK, Günaydın M. Cepheid Xpert vanA/vanB testinin rektal örneklerde vankomisin-dirençli enterokokların hızlı tespiti için değerlendirilmesi. 4th EKMUD Congress (Infectious Diseases & Clinical Microbiology); 9–12 May 2012; Pendik, İstanbul, Turkey. pp. 189.

30. Durmaz S, Erçal BD, Atalay A, Perçin D. Vankomisine dirençli enterokok saptanmasında GeneXpert vanA/vanB real time-PCR yöntemi ile kültür yönteminin karşılaştırılması. XXXIV. Türk Mikrobiyoloji Kongresi; 7–11 Kasım 2010; Girne, Kuzey Kıbrıs Türk Cumhuriyeti. pp. 237.