Vancomycin-Resistant Enterococci (VRE) Isolates Isolated in Rajavithi Hospital between 1999 and 2009

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From 1999 to 2009, a total of 10,470 clinical enterococcal strains from patients in Rajavithi Hospital were isolated. Of these, 201 (1.9%) vancomycin-resistant enterococci (VRE) including 199 (99.0%) Enterococcus faecium and 2 (1.0%) Enterococcus faecalis were found. The incidence of VRE was 1.8% in 1999, 3.3% in 2000, 5.1% in 2001, 1.0% in 2002, 0.0% in 2003 and 2004, 1.1% in 2005, 4.7% in 2006, 1.9% in 2007, 2.2% in 2008 and 0.9% in 2009. Seventy-one VRE isolates were classified to VanA phenotype (69 VanA E. faecium and 2 VanA E. faecalis) and 130 were classified to VanB phenotype (VanB E. faecium). The rate of inpatient departments (IPD)-associated VRE (199 (99.0%) VRE isolates) was significantly greater than the incidence of outpatient departments (OPD)-VRE (2 (1.0%) VRE isolates). VRE were found in medical (33.7%), ICUs (15.6%) and surgical (15.1%) wards. VRE were mostly found in urine, 64.2%, following in pus, blood, genital and sputum specimens, 21.9%, 9.0%, 3.5% and 1.4%, respectively. VRE, especially vancomycin resistant E. faecium, were multidrug-resistant (resistance to ampicillin, tetracycline, norfloxacin, ciprofloxacin, erythromycin and gentamicin). All strains of VRE were fully susceptible to linezolid.

Keywords: *Vancomycin-resistant Enterococci, VRE, Enterococcus faecalis, Enterococcus faecium, Antimicrobial susceptibility testing*

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Since the first report of vancomycin-resistant enterococci (VRE) in 1986[1], VRE have become increasingly nosocomial pathogens worldwide[2]. However, the epidemiology of VRE varies widely in different geographical areas[3]. Infections with VRE have been increasingly severe for enterococcal treatment since they can transfer antibiotic resistance genes to *Staphylococcus aureus*, especially methicillin resistant *Staphylococcus aureus* (MRSA)[2,4]. So, rapid and accurate identification of VRE is critical in management and treatment to limit spread of VRE[5]. There are six glycopeptide resistance phenotypes (VanA to VanE and VanG). They can be distinguished on the basis of the sequence of the structural gene for the resistance ligase *(vanA to vanE and vanG)*[6]. All phenotypes except for VanC are corresponded to acquired resistance to glycopeptides and transferable. The VanA and VanB phenotypes are the most important pathogens. Both phenotypes have been mostly found in *E. faecium* and *E. faecalis* which are major causes of human pathogens. VanA phenotype is characterized by high-level resistance to both vancomycin and teicoplanin, whereas VanB phenotype is characterized by variable resistance to vancomycin, but susceptible to teicoplanin. In Thailand, the first report of VRE isolate was from King Chulalongkorn Memorial Hospital [7]. The data of VRE in Thailand is little known. The aims of the present study were to characterize the phenotypes and to evaluate the prevalence of VRE in Rajavithi Hospital during 1999 to 2009, including sites of infection, wards and resistance patterns. All data were analyzed and presented in term of frequency and percentage distribution during 1999 to 2009.

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Material and Method

Bacterial strains

All clinical enterococcal isolates recovered from all specimens (except stool) were studied between January 1999 and December 2009 in Rajavithi Hospital, a 1,200-bed teaching institute. In VRE screening, enterococcal isolates with vancomycin zone of inhibition of less than or equal to 16 mm in diameter

were selected[8]. All VRE isolates were collected and stored at -70°C. *E. faecalis* ATCC29212 (vancomycin-susceptible strain), *E. faecalis* ATCC51299 (*vanB* gene), *E. faecium* UCLA 192 (*vanB* gene), *E. gallinarum* ATCC49573 (*vanC1* gene) and *E. casseliflavus* ATCC25788 (*vanC2* gene) were used as control strains.

Phenotypic species identification

All enterococcal strains were identified by conventional methods[9]. Before 1999, enterococcal strains were only identified to genus level based on esculin hydrolysis and pyrrolidonylarylamidase (PYR) activities, indicating genus streptococci group D, enterococci. Since 2000, all enterococcal strains have been reported at species level as *E. faecalis*, *E. faecium* and *Enterococcus* species (spp.), on the basis of growth on tellurite agar, dihydrolysis of arginine, fermentation of mannitol, arabinose, methyl-alpha-D-glucopyranoside and pyuruvate.

Phenotypic characterization of VRE isolates

To distinguish Van phenotype, minimal inhibitory concentrations (MICs) of vancomycin and teicoplanin were performed by using Mueller-Hinton agar according to CLSI guidelines[8]. *E. faecalis* or *E. faecium* isolates were defined as having the VanA phenotype if the vancomycin MIC was greater than 32 microgram/millitre (mcg/ml) and the teicoplanin MIC was greater than 32 mcg/ml. These two species were defined as having the VanB phenotype if the vancomycin MIC was greater than 4 mcg/ml and the teicoplanin MIC was less than 8 mcg/ml[6,8].

Species identification and genotypic characterization of VRE isolates by polymerase chain reaction (PCR)

Seventy and six of VRE isolates including 33 VanA *E. faecium*, 2 VanA *E. faecalis* which isolated between 1999 and 2002 and 35 VanB *E. faecium*, 3 VanC *E. gallinarum* and 3 VanC *E. casseliflavus* which isolated between 2005 and 2009 were confirmed species identification and vancomycin resistance genotype by PCR. Enterococci were grown overnight at 37°C in Todd-Hewitt broth and then 1 ml volumes were centrifuged at 10,000 rpm, 1 min. The pellet was resuspended in 200 microlitre (mcl) of TE buffer (10 mM Tris-HCl, 1 mMEDTA pH 8.0), heated at 95°C for 20 min and centrifuged at 10,000 rpm, 2 min. Primer sequences of each gene are summarized in Table 1. Five microlitre volumes of the supernatant were subjected to PCR amplification in 25 mcl reaction mixtures containing each primers (3.0 pmol of *vanC1*, *E. faecium*, 16S rRNA (*rrs*); 6.5 pmol of *vanA*, *vanC2/C3*, *E. faecalis*[10]; 12.5 pmol of *vanB*[11], 200 mcM each deoxynucleotide triphosphate, 10 mM Tris HCl pH 8.3, 50 mM KCl, 1.5 mM $MgCl_2$ and 1 U of Taq polymerase (Quiagen, Germany)[10]. DNA amplification was carried out with the following conditions: 94°C, 5 min, 30 cycles of amplification (94°C, 1 min, 54°C, 1 min, 72°C, 1 min) and a final extension at 72°C for 10 min. PCR products were analyzed on a 1.5% Seakem ME agarose gel (FMC Bioproducts, Rockland, Maine). A 100-basepairs (bp) was used as molecular size marker. The gels were strained with Ethidium bromide and photographed under UV light. PCR products with sizes of 1,030, 635, 941, 658, 822, 484 and 320 bp were corresponded to the

Table 1. Oligonucleotide primers and products of vancomycin-resistant enterococci

Ampliflied gene	Primer pair sequence	Size of PCR product (bp)	Ref
vanA	CATGAATAGAATAAAAGTTGCAATA CCCCTTTAACGCTA	1,030	10
vanB	ATGGGAAAGCCGATAGTC GATTTCGTTCCTCGACC	635	11
vanC1	GGTATCAAGGAAACCTC CTTCCGCCATCATAGCT	822	10
vanC2/C3	CGGGGAAGATGGCAGTAT CGCAGGGACGGTGATTTT	484	10
E. faecalis	ATCAAGTACAGTTAGTCTTTATTAG ACGATTCAAAGCTAACTGAATCAGT	941	10
E. faecium	TTGAGGCAGACCAGATTGACG TATGACAGCGACTCCGATTCC	658	10
rrs (16S rRNA)	GGATTAGATACCCTGGTAGTCC TCGTTGCGGGACTTAACCCAAC	320	10

vanA, vanB, E. faecalis, E. faecium, E. gallinarum, E. casseliflavus and *rrs* target, respectively.

Antimicrobial susceptibility testing

Antibiotic resistance was performed by disk diffusion method on Mueller-Hinton agar according to CLSI guidelines[8] for the following antimicrobial agents: ampicillin (10 mcg), gentamicin (120 mcg), tetracycline (10 mcg), vancomycin (30 mcg), teicoplanin (30 mcg) and linezolid (30 mcg). The MIC for vancomycin and teicoplanin was determined by E test. *S. aureus* ATCC25923 and *E. faecalis* ATCC29212 were used as control strains.

This research was approved by the research ethics board of Rajavithi hospital.

Results

Vancomycin-resistant enterococci (VRE) isolates

Between 1999 and 2009, a total of 10,470 clinical enterococcal isolates, including 964 streptococci group D, enterococci, 4,827 *E. faecalis*, 4,106 *E. faecium* and 573 *Enterococcus* spp., were isolated (data not shown). Of these, 201 (1.9%) of VRE were found. The incidence of VRE isolates was 12 (1.8%) in 1999, 21 (3.3%) in 2000, 32 (5.1%) in 2001, 6 (1.0%) in 2002, 0 (0.0%) in 2003 and 2004, 12 (1.1%) in 2005, 54 (4.7%) in 2006, 25 (1.9%) in 2007, 28 (2.2%) in 2008 and 11 (0.9%) in 2009 (Fig. 1 and Table 2). Among 201 VRE isolates, 199 (99.0%) were *E. faecium* (vancomycin-resistant *E. faecium*) and 2 (1.0%) were *E. faecalis* (vancomycin-resistant *E. faecalis*) (Table 2). Both vancomycin-resistant *E. faecalis* strains were found in 2001 and 2002 (Table 5).

Phenotypic characterization of VRE isolates by E test

Of the 201 VRE isolates, 71 had VanA phenotype and 130 had VanB phenotype. Between 1999 and 2002, all of VRE isolates (71) had VanA phenotype including 69 VanA *E. faecium* and 2 VanA *E. faecalis*. Between 2005 and 2009, all of VRE isolates (130) had VanB *E. faecium* (Table 2). All VanA *E. faecium* isolates (69) had high MICs for vancomycin (greater than 256 mcg/ml) and teicoplanin (8-64 mcg/ml). Most of them (51.6%) with an MIC of teicoplanin 32 mcg/ml were isolated. One hundred and thirty of VanB *E. faecium* isolates were resistant to vancomycin (MICs, greater than or equal to 32 mcg/ml), but were susceptible to

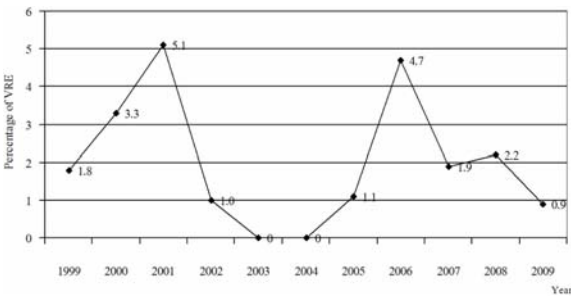

Fig. 1 Vancomycin resistance among enterococci isolated in Rajavithi Hospital between 1999 and 2009

Table 2. Numbers (%) of VRE and numbers of VanA and VanB phenotypes in *E. faecium* and *E. faecalis* isolates isolated in Rajavithi Hospital, 1999-2009. (n=201)

Year	Total Enterococcal isolates	Total VRE isolates	(%)	Van Phenotypes			
				VanA		VanB	
				E. faecium	*E. faecalis*	*E. faecium*	*E. faecalis*
1999	657	12	1.8	12	-	-	-
2000	644	21	3.3	21	-	-	-
2001	630	32	5.1	31	1	-	-
2002	604	6	1.0	5	1	-	-
2003	800	-	-	-	-	-	-
2004	981	-	-	-	-	-	-
2005	1,142	12	1.1	-	-	12	-
2006	1,153	54	4.7	-	-	54	-
2007	1,296	25	1.9	-	-	25	-
2008	1,284	28	2.2	-	-	28	-
2009	1,280	11	0.9	-	-	11	-
Total	10,470	201	1.9	69	2	130	-

teicoplanin (MICs, 0.25-2.0 mcg/ml). Most of them, 62.3%, showed high-level resistance to vancomycin (MICs, greater than 256 mcg/ml) and 36.8%, showed susceptible to teicoplanin (MIC, 0.5 mcg/ml). Both VanA *E. faecalis* isolates had high MICs for vancomycin (MICs, greater than 256 mcg/ml) and teicoplanin (MICs, 8 and 32 mcg/ml) (data not shown).

Species identification and genotypic characterization of VRE isolates by PCR

As shown in Fig. 2, PCR products with a size of 320 bp corresponding to the *rrs* target (internal control) were observed in all of the isolates (lane 1 to 10). The internal PCR control supplied guarantee that the clinical specimens were successfully amplified and detected. As shown in Fig. 2, one band with size of 941, 658 and 635 bp were produced from an *E. faecalis*-specific (lane 1), *E. faecium*-specific (lane 2 and 6) and the *vanB* (lane 7) target, respectively. Two bands with sizes of 1,030 and 941 bp corresponded to the *vanA* and *E. faecalis*-specific targets (lane 3) and two of 635 and 941 bp corresponded to the *vanB* and *E. faecalis*-specific targets (lane 4), indicating they were products from an *E. faecalis vanA* isolate and an *E. faecalis vanB* isolate, respectively. Two bands with sizes of 1,030 and 658 bp corresponded to the *vanA* and *E. faecium*-specific targets (lane 5) and two of 635 and 658 bp corresponded to the *vanB* and *E. faecium*-specific targets (lane 6), indicating they were products from an *E. faecium vanA* isolate and an *E. faecium vanB* isolate, respectively. One band with a size of 822 bp (lane 8) or 484 bp (lane 9) corresponded to the *vanC1* (*E. gallinarum*) or *vanC2* (*E. casseliflavus*) target, respectively. Of 76 isolates tested, 100% of the expected PCR products with intense bands on the gel were produced. Two bands with sizes of 1,030 and 658 bp corresponding to the *vanA* and *E. faecium*-specific targets were produced from all (33 isolates) VanA *E. faecium* isolates. Two bands with sizes of 635 and 658 bp corresponding to the *vanB* and *E. faecium*-specific targets were produced from all (35 isolates) VanB *E. faecium* isolates (Fig. not shown). The correlation between the phenotypic and genotypic methods in the species identification and van-type characterization was 100% in the present study (Table 3).

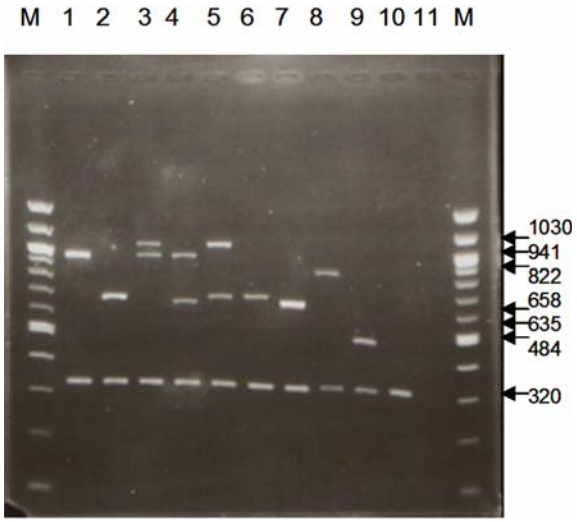

Fig. 2 Agarose gel electrophoresis of amplified *E. faecalis*-specific, *E. faecium*-specific, *vanA, vanB, vanC1, vanC2/C3* and *rrs* genes by the multiplex PCR assays containing seven primers sets. Lanes: M; 100-bp DNA ladder (New England Biolabs); 1, an *E. faecalis* (941 bp) isolate; 2, an *E. faecium* (658 bp) isolate; 3, an *E. faecalis vanA* (941 and 1,030 bp) isolate; 4, an *E. faecalis vanB* (941 and 635 bp) isolate; 5, an *E. faecium vanA* (658 and 1,030 bp) isolate; 6, an *E. faecium vanB* isolate (658 bp for *E. faecium* band); 7, an *E. faecium vanB* isolate (635 bp for *vanB* band); 8, an *E. gallinarum vanC1* (822 bp) isolate; 9, an *E. casseliflavus vanC2/3* (484 bp) isolate; 1-10, 16S rRNA (*rrs*) (320 bp)

Table 3. Phenotypic and genotypic characterization of Van type of 76 VRE isolates isolated in Rajavithi Hospital between 1999 and 2007

Species	No. of isolates	Vancomycin disk zone of inhibition (mm)	MICs of (mcg/ml)		Van Phenotype	*van* Genotype
			Vancomycin	Teicoplanin		
E. faecium	33	6	greater than 256	greater than 48	VanA	*vanA*
E. faecalis	2	6	greater than 256	8, 32	VanA	*vanA*
E. faecium	35	6	48-greater than 256	0.32-2.0	VanB	*vanB*
E. faecalis	-	-	-	-	-	-
E. gallinarum	3	17	6-8	0.75	VanC	*vanC1*
E. casseliflavus	3	17	4-6	0.75-1	VanC	*vanC2*

Sources of VRE isolates

Of 201 VRE isolates, 2 (1.0%) isolates were found in outpatient departments (OPD) and 199 (99.0%) isolates in inpatient departments (IPD). Among 199 of IPD-associated VRE isolates, 67 (33.7%) were from medical wards, 31 (15.6%) were from intensive care units (ICU) wards, 30 (15.1%) were from surgical wards, 10 (5.0%) were from gynecology wards and 61 (30.6%) were from the other wards (Table 4). Between 1999 and 2009, occurrence of VRE rate in each ward was steady. Of 201 VRE isolates, most were frequently found in urine (129, 64.2%), following by pus (44, 21.9%), blood (18, 9.0%), genital (7, 3.5%) and sputum (3, 1.4%) specimens (Table 5). Both VREfaecalis were isolated from urine and blood, respectively. No VRE isolate was found from cerebrospinal fluid (CSF) in this present study. Over an 11-year period, occurrence of VRE rate in each specimen was steady.

Antimicrobial susceptibility testing

Susceptibility to various antibiotics of *E. faecalis* and *E. faecium* was as follows: ampicillin 99.0% and 4.0%, high level gentamicin 60.0% and 26.0%, tetracycline 14.0% and 9.0%, chloramphenicol 61.0% and 90.0%, erythromycin 20.0% and 5.0% and vancomycin resistance in 99.9% and 94.8% respectively. All enterococcal strains were susceptible to linezolid and teicoplanin, 100% and 99.9%, respectively (data not shown).

The susceptibility of VRE to these antibiotics are shown in Table 6; in VREfaecium isolates, only chloramphenicol, teicoplanin and linezolid showed

Table 4. Occurrence of 201 VRE isolates isolated from OPD and IPD wards in Rajavithi Hospital between 1999 and 2009

Year	Total	OPD	IPD	IPD				
				ICU	Sur	Med	Gyn	Others
1999	12	-	12	3	-	6	1	2
2000	21	1	20	2	-	13	-	5
2001	32	-	32	10	7	10	-	5
2002	6	1	5	-		1	-	4
2005	12	-	12	4	1	2	1	4
2006	54	-	54	5	9	17	4	19
2007	25	-	25	3	2	9	3	8
2008	28	-	28	1	7	8	1	11
2009	11	-	11	3	4	1	-	3
Total (%)	201	2 (1.0)	199 (99.0)	31 (15.6)	30 (15.1)	67 (33.7)	10 (5.0)	61 (30.6)

Table 5. Occurrence of 201 VRE isolates isolated from various specimens in Rajavithi Hospital between 1999 and 2009

Year	VRE strains	Total	No. of isolates per site infection				
			Blood	Urine	Sputum	Genital	Pus
1999	*E. faecium*	12	2	4	-	-	6
2000	*E. faecium*	21	-	15	-	1	5
2001	*E. faecium*	31	-	21	-	1	9
	E. faecalis	1	1	-	-	-	-
2002	*E. faecium*	5	-	4	1	-	-
	E. faecalis	1	-	1	-	-	-
2005	*E. faecium*	12	2	7	-	2	1
2006	*E. faecium*	54	6	41	1	2	4
2007	*E. faecium*	25	2	13	-	1	9
2008	*E. faecium*	28	2	17	1	-	8
2009	*E. faecium*	11	3	6	-	-	2
Total (%)		201	18 (9.0)	129 (64.2)	3 (1.4)	7 (3.5)	44 (21.9)

Table 6. Percentage of susceptibility of 10 antimicrobial agents to 201 VRE isolates isolated in Rajavithi Hospital between 1999 and 2009

Antimicrobial agents	All VRE Strains		VRE in Blood		VRE in Urine	
	E. faecium	*E. faecalis*	*E. faecium*	*E. faecalis*	*E. faecium*	*E. faecalis*
Ampicillin	0 (167)	100.0 (2)	0 (11)	100.0 (1)	0 (100)	100.0 (1)
Gentamicin	20.7 (58)	100.0 (1)	0 (10)	100.0 (1)	28.6 (7)	-
Norfloxacin	0 (24)	-	-	-	0 (24)	-
Ciprofloxacin	0 (42)	0 (1)	-	-	0 (42)	0 (1)
Tetracycline	5.2 (154)	0 (1)	9.1 (11)	-	4.6 (110)	0 (1)
Chloramphenicol	100.0 (25)	-	100.0 (3)	-	-	-
Erythromycin	1.9 (54)	-	0 (11)	-	-	-
Vancomycin	0 (167)	0 (2)	0 (11)	0 (1)	0 (110)	0 (1)
Teicoplanin	99.0* (113)	0 (1)	100.0 (11)	-	98.7* (75)	0 (1)
Linezolid	100 (91)	-	100.0 (11)	-	100.0 (60)	-

(No. of tests), * VanB VRE isolates were 100% susceptible to teicoplanin

good activity (99.0-100%). The sensitivity of VRE isolated from blood was susceptible to chloramphenicol, teicoplanin and linezolid, but those from urine were only susceptible to teicoplanin and linezolid. VREfaecalis isolates were susceptible to ampicillin and gentamicin, but resistant to ciprofloxacin, tetracycline, vancomycin and teicoplanin.

Discussion

Over an 11-year period during 1999 to 2009, the rate of incidence of VRE in Rajavithi Hospital varied from lowest, 0.0% in 2003 and 2004 to highest, 4.7% and 5.1% in 2006 and 2001, respectively. The trend of VRE findings in Rajavithi Hospital could not be exactly predicted but it could indicate the low prevalence. However, from the present study, the prevalence of VRE in Rajavithi Hospital showed critical trend because the ratio of clinical *E. faecalis* to *E. faecium* was 5:4. In another study, Facklam RR et al[12] reported a different ratio at 9:1. The present study reported that VRE isolates were mostly found as *E. faecium* (99%) and a lot less as *E. faecalis* (1%), while the study of Edmond MB et al[13] reported different rate of VRE, *E. faecium* (50.5%) and *E. faecalis* (3.1%).

VRE isolated found during the period from 1999 to 2002 were VanA-type only. In contrast, from 2005 to 2009, there was only VanB-type of VRE isolates. This phenomenon could not be explained by the authors. Notably, VanB *E. faecium* strains were still found in Rajavithi Hospital during 2010 as well as in King Chulalongkorn Memorial Hospital[7]. However, the epidemiology of VRE types (VanA or VanB) varies widely in different geographic areas. For example, VanA are mostly found in the United States, while VanB are mostly found in Europe[3].

The present study compared the correlation of phenotypic and genotypic characterization using E-test and PCR, respectively. The susceptibility and the sensitivity levels were 100%. This is because all strains of VRE had an inhibition zone for vancomycin disk 6 mm. In addition, most of them had high level resistance. VanA-type were resistant to vancomycin (MICs, greater than 256 mcg/ml) and teicoplanin (MICs, 32-64 mcg/ml) while VanB-type were susceptible to teicoplanin (MIC, 0.5 mcg/ml), giving the ability to classify into VanA or VanB phenotype. Only one isolate of *E. faecalis* was resistant to vancomycin (MIC, greater than 256 mcg/ml), but was moderately susceptible to teicoplanin (MIC, 8 mcg/ml). Therefore, it was classified into VanA-type, but might be VanA or VanB phenotype. PCR testing eventually confirmed that this organism had *vanA* genotype. Classification of Van phenotypes using MICs of vancomycin and teicoplanin had limitation in case of low level of glycopeptide resistance[14]. For example, some VanB phenotypes (susceptible to teicoplanin) had *vanA* genotype[15]. In contrast, Vandamme P et al[16] reported that VanA phenotype (resistant to vancomycin and teicoplanin) strain had *vanB* genotype. So, the correction of genotypic characterization of *van* needs to be confirmed by genotypic methods.

Most of VRE were major causes of nosocomial pathogens. Most of VRE in Rajavithi Hospital were isolated from IPD, in medical wards, while the study of Bonadio M et al[17] reported that VRE were isolated more from OPD than that from IPD. Some studies also

reported that VRE were isolated from community-acquired infections[18,19]. Most of VRE in Rajavithi Hospital were found from urine specimens, like that reported by Bonadio M et al[17]. Nevertheless, Low DE, et al[3] reported that VRE were found mostly in blood specimens.

The prevalence rate of VRE in Rajavithi Hospital was 1.9%, which was higher than that in King Chulalongkorn Memorial Hospital (0.8%)[7]. However, this rate (1.9%) could be considered relatively low when comparing with the rates of VRE in Italy, USA and United Kingdom, which were 19.6%, 14.8% and 10.4%, respectively[20,21]. The studies in Autralia, Belgium and Netherlands found no VRE isolates[20]. In Asia, there are several reports describing the isolation of VRE, including India and Taiwan[22,23].

Generally, *E. faecium* had higher multidrug resistance characteristics than that of *E. faecalis*. Especially, vancomycin-resistant *E. faecium* is resistant to ampicillin, high-level gentamicin, ciprofloxacin, levofloxacin, erythromycin, vancomycin and teicoplanin (in case of VanA), but is susceptible to chloramphenicol. Unfortunately, most VRE were found in urine, which could not be treated by chloramphenicol. The drug of choice for VRE treatment was linezolid[24]. Recently, linezolid-resistant *E. faecium*[25] clinical isolates have been reported.

Conclusion

Although the incidence rate of VRE in Rajavithi Hospital was still low, the present study showed the serious problem of further spread of VRE. The supporting ideas are as the following. First, *E. faecium*, the commonly multidrug resistance strain causing difficult treatment, was isolated as many as *E. faecalis*. Second, most of VRE isolated in Rajavithi Hospital were VanA and VanB phenotypes that were mostly found in *E. faecium* and *E. faecalis*. These two phenotypes were able to transfer resistance gene to other gram positive bacteria, especially MRSA. Last, in Rajavithi Hospital, 50.0% of *S. aureus* was MRSA and drug of choice of MRSA is vancomycin. So, it might become a serious problem if these VanA and VanB VRE isolated have transferred *van* gene to other enterococci, *E. faecalis* and *E. faecium* and MRSA. Finally, this problem requires the supplement of infectious control team to limit further spread.

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Potential conflicts of interest

None.

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เชื้อ Enterococci ดื้อต่อยา vancomycin (VRE) ที่แยกได้จากโรงพยาบาลราชวิถี ระหว่างปี พ.ศ. 2542-2552

ประกายทิพย์ ทองคุ้ม, สุวัฒนา กาญจนหฤทัย, สุริวรรณ จันทรคุปตังกูร, สมศักดิ์ ราหุล

ตั้งแต่ปี พ.ศ. 2542 ถึง 2552 ทำการแยกเชื้อ enterococci ก่อโรคจากสิ่งส่งตรวจของผู้ป่วยโรงพยาบาลราชวิถี ทั้งหมดจำนวน 10,470 สายพันธุ์ พบเชื้อ enterococci ดื้อต่อยา vancomycin (VRE) จำนวน 201 สายพันธุ์ คิดเป็นร้อยละ 1.9 ประกอบด้วยเชื้อ Enterococcus faecium จำนวน 199 สายพันธุ์ (ร้อยละ 99.0) และเชื้อ Enterococcus faecalis จำนวน 2 สายพันธุ์ (ร้อยละ 1.0) อัตราการพบเชื้อ VRE พบร้อยละ 1.8 (พ.ศ. 2542), ร้อยละ 3.3 (พ.ศ. 2543), ร้อยละ 5.1 (พ.ศ. 2544), ร้อยละ 1.0 (พ.ศ. 2545), ร้อยละ 0 (พ.ศ. 2546, พ.ศ. 2547), ร้อยละ 1.1 (พ.ศ. 2548), ร้อยละ 4.7 (พ.ศ. 2549), ร้อยละ 1.9 (พ.ศ. 2550), ร้อยละ 2.2 (พ.ศ. 2551) และ ร้อยละ 0.9 (พ.ศ. 2552) เชื้อ VRE เป็นชนิด VanA จำนวน 71 สายพันธุ์ (เชื้อ VanA E. faecium 69 สายพันธุ์ และ เชื้อ VanA E. faecalis 2 สายพันธุ์) และเป็นชนิด VanB จำนวน 130 สายพันธุ์ (เชื้อ VanB E. faecium) เชื้อ VRE ที่แยกได้จากผู้ป่วยใน (IPD) จำนวน 199 สายพันธุ์ (ร้อยละ 99.0) มากกว่าเชื้อที่แยกได้จากผู้ป่วยนอก (OPD) จำนวน 2 สายพันธุ์ (ร้อยละ 1.0) อย่างมีนัยสำคัญ เชื้อ VRE ตรวจพบจากตึกอายุรกรรม ตึกหออภิบาลผู้ป่วยและตึกศัลยศาสตร์ คิดเป็นร้อยละ 33.7, 15.6 และ 15.1 ตามลำดับ เชื้อ VRE พบมากที่สุดในสิ่งส่งตรวจประเภทปัสสาวะคิดเป็นร้อยละ 64.2 รองลงมาพบในสิ่งส่งตรวจประเภทแผลหนอง, เลือด, ระบบอวัยวะสืบพันธุ์ และเสมหะ คิดเป็นร้อยละ 21.9, 9.0, 3.5 และ 1.4 ตามลำดับ เชื้อ VRE โดยเฉพาะอย่างยิ่ง เชื้อ VREfaecium ดื้อต่อยาหลายชนิด (ดื้อต่อยา ampicillin, ยา tetracycline, ยา ciprofloxacin, ยา norfloxacin, ยา erythromycin และ ยา gentamicin) เชื้อ VRE ทุกสายพันธุ์ไวต่อยา linezolid