Original Article

Vancomycin-resistant Enterococci in Singaporean Hospitals: 5-year results of a Multi-centre Surveillance Programme

Yiying Cai,[1] *BSc (Pharm)*, Joey PJ Chan,[2] *FRCPath*, Dale Andrew Fisher,[3] *FRACP*, Li Yang Hsu,[3] *MPH*, Tse Hsien Koh,[4] *FRCPath*, Prabha Krishnan,[5] *FRCPath*, Andrea LH Kwa,[1] *PharmD*, Thean Yen Tan,[6] *MRCPath*, Nancy WS Tee,[7] *FRCPA*

Abstract

Introduction: Vancomycin-resistant enterococci (VRE) have emerged as one of the major nosocomial antimicrobial-resistant pathogens globally. In this article, we describe the epidemiology of VRE in Singaporean public hospitals in the 5 years following the major local VRE outbreak in 2005. Materials and Methods: A passive laboratory surveillance programme identified non-duplicate VRE isolates from 7 hospitals from 2006 to 2010. Descriptive statistics and time-series analysis was performed on all clinical VRE isolates for each individual hospital as well as for the combined dataset. Results: There were a total of 418 VRE isolates over 5 years, of which 102 isolates (24.4%) were from clinical cultures. Between 0.4% and 0.7% of all clinical enterococcal isolates were resistant to vancomycin. The overall incidence-density of VRE did not change over time in Singapore despite 2 separate outbreaks in tertiary hospitals in 2009 and 2010. Incidence-density of clinical VRE cases fell in 2 secondary hospitals, while another 2 hospitals experienced no significant VRE infections after 2008. Conclusion: The prevalence of VRE clinical isolates remains low in Singaporean public sector hospitals. However, the presence of at least 2 outbreaks in separate hospitals over the past 5 years indicates the need for continued vigilance in order to prevent any further increase in VRE prevalence locally.

Ann Acad Med Singapore 2012;41:77-81

Key words: Vancomycin-resistant enterococci, Passive surveillance, Antimicrobial resistance, endemics

Introduction

Vancomycin-resistant enterococci (VRE) rapidly became a significant cause of nosocomial infections in many modern hospitals worldwide following its emergence in 1986 in UK and France.[1,2] Although of lower virulence compared to other common nosocomial pathogens, these hardy organisms can cause serious and life-threatening infections, especially in immunocompromised patients.[3] In 2004, VRE were classified along with 5 other groups of drug-resistant microbes on the Infectious Diseases Society of America's list of global "bad bugs" for which the development of new drugs was urgently required, comprising more than 27% of enterococci from clinical samples in American intensive care units in 2002.[4]

Two VRE outbreaks have been reported in Singaporean hospitals to date.[5] Both were caused by *van*B-carrying *Enterococcus faecium* and originated in Singapore's largest hospital, with the second outbreak involving multiple local hospitals before it was finally controlled in mid-2005.[6]

The Network for Antimicrobial Resistance Surveillance (Singapore) (NARSS) was established in December 2005 to conduct prospective laboratory- and pharmacy-based surveillance of antibiotic resistance and prescription in

[1]Department of Pharmacy, Singapore General Hospital, Singapore
[2]Department of Laboratory Medicine, Khoo Teck Puat Hospital, Singapore
[3]Department of Medicine, National University Health System, Singapore
[4]Department of Pathology, Singapore General Hospital, Singapore
[5]Department of Laboratory Medicine, Tan Tock Seng Hospital, Singapore
[6]Department of Laboratory Medicine, Changi General Hospital, Singapore
[7]Department of Laboratory, KK Hospital, Singapore
Address for Correspondence: Dr Hsu Li Yang, National University Health System, 1E Kent Ridge Road, NUHS Tower Block Level 10, Singapore 119228.
Email: mdchly@nus.edu.sg

local public sector hospitals. The objective of this study was to analyse and describe the annual trend of VRE in local hospitals in the years from 2006 to 2010, 5 years after the last major outbreak.

Materials and Methods

Participating Hospitals and Period

Seven hospitals participated in this study. Hospitals 1, 2, 4 and 7 are secondary hospitals – Hospital 1 closed in early 2010 while Hospital 4 started its inpatient service shortly afterwards. Hospitals 5 and 6 are tertiary hospitals while Hospital 3 offers only paediatrics and obstetrics/gynaecological services.

The period of surveillance was from January 2006 to December 2010. Hospital 1 provided data until December 2009 whereas Hospital 4 provided data only for the second half of 2010. Complete datasets covering the entire surveillance period were obtained from all other hospitals. In general, active screening for VRE among inpatients was not conducted at any hospital except during outbreaks, where there was an increase in isolates obtained from clinical specimens.

Data Collection

Microbiologic data were extracted from the laboratory information system of each hospital and converted centrally into a standard format using WHONET 5.5 (WHO, Geneva, Switzerland), with duplicates eliminated on an annual basis according to the guidelines of the Clinical and Laboratory Standards Institute (CLSI).[7] Infection control staff provided the data for Hospital 5; laboratory staff from all other hospitals provided the microbiologic data directly. Identification and susceptibility testing of VRE were performed at the level of the contributing laboratories. VRE were segregated into clinical and screening isolates based on culture specimens – isolates cultured from stool and rectal swabs were considered screening isolates whereas those cultured from all other patient sites were classified as clinical isolates. Denominator data in the form of hospital inpatient-days were obtained from the hospitals' administrative records.

Statistical Analysis

Each individual hospital and the combined hospital VRE results were expressed as incidence-density per 10,000 inpatient-days respectively for each year. These series were then explored independently for trend over time by linear

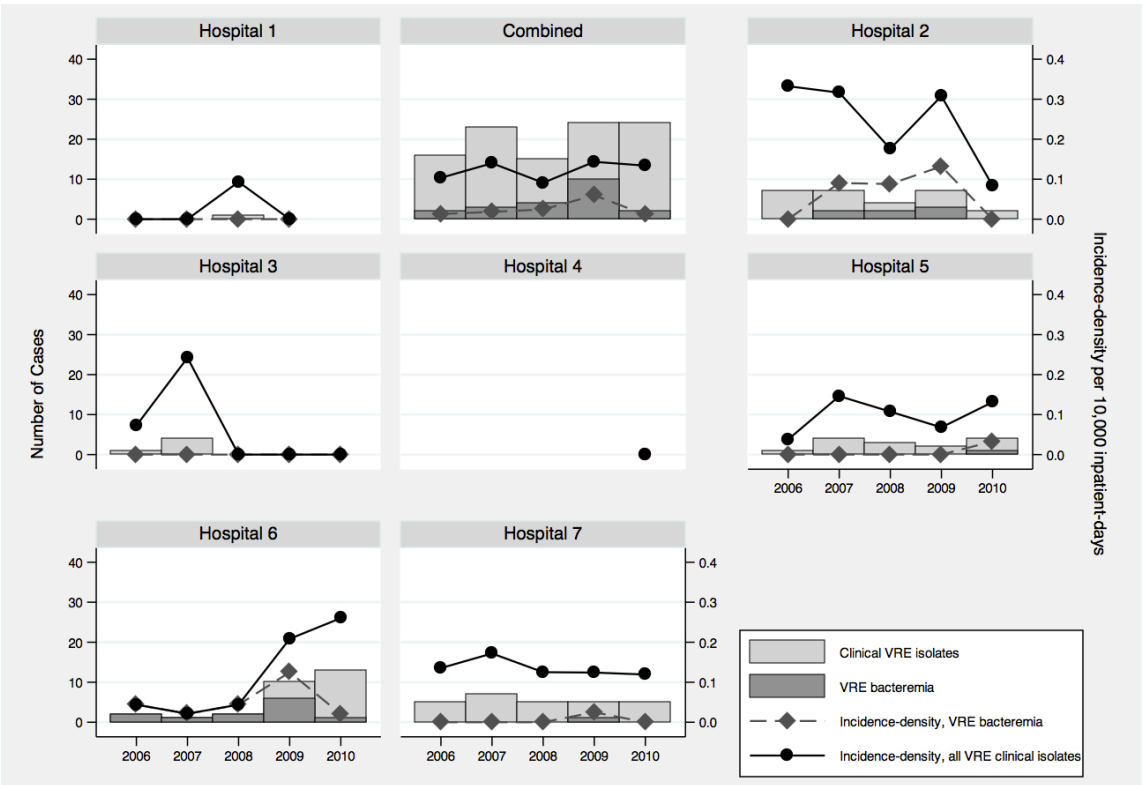

Fig. 1. Incidence-density and number of vancomycin-resistant enterococci isolates from clinical and blood cultures, by hospital, 2006-2010.

Table 1. Number of vancomycin-resistant enterococci (VRE) and percentage of vancomycin resistance among all *Enterococcus* spp. isolates in Singaporean hospitals.

Year	All VRE isolates, number (range*)	Percentage vancomycin resistance^ (range*)	Clinical VRE isolates, number (range*)	Percentage clinical vancomycin resistance^ (range*)
2006	57 (0-20)	1.5% (0%-6.9%)	15 (0-7)	0.4% (0%-1.5%)
2007	78 (0-26)	1.6% (0%-7.1%)	19 (0-7)	0.6% (0%-2.5%)
2008	71 (0-23)	1.5% (0%-6.2%)	12 (0-5)	0.4% (0%-1.8%)
2009	114 (0-74)	3.3% (0%-4.5%)	22 (0-10)	0.7% (0%-2.3%)
2010	98 (0-41)	2.3% (0%-6.6%)	20 (0-13)	0.7% (0%-2.4%)

* Distribution range among participating hospitals.
^ Calculated using the number of all *Enterococcus* spp. cultured as the denominator. Results from Hospital 5 were not included as the denominator figures were not available.

regression, corrected for first-level autocorrelation using the Cochrane-Orcutt estimation following determination of the Durbin-Watson statistic. A coefficient of determination (R^2) of >0.3 coupled with $P \leq 0.05$ was considered to be a statistically significant trend result. Statistical analysis was performed using Stata 11.1 (Statacorp, Texas, USA).

Results

There were 418 non-duplicate VRE isolates cultured over the 5-year period, of which 102 (24.4%) were from clinical specimens. Of the latter, 21 (20.6%) were obtained from blood cultures, while the majority (91.4%) of the remaining isolates was from urine cultures. These isolates included *E. faecalis*, *E. faecium* as well as other *Enterococcus* spp. reported by the contributing laboratory as being resistant to vancomycin. *E. faecium* was the most prevalent, representing between 78.9% to 91.8% of VRE isolated over 5 years, whereas non *E. faecalis/faecium* isolates comprised <2% of all VRE isolated. The total number of VRE isolated in the study period along with the percentage of vancomycin resistance is shown in Table 1, whereas the number and incidence-density of clinical VRE isolates by hospital is shown in Figure 1. None of the VRE isolated in Hospitals 1 and 3 were *E. faecalis* or *E. faecium*.

The increased number of all (screening plus clinical) VRE isolates in 2009 and 2010 were due to outbreaks in the 2 tertiary hospitals (Hospitals 6 and 5 respectively). Hospital 6 saw a threefold increase in incidence-density of all VRE isolates within a year to 1.54 cases per 10,000 inpatient-days in 2009, whereas a similar increase was seen in Hospital 5 (to 1.21 cases per 10,000 inpatient-days) in 2010. Overall, however, there was no statistically significant increase/change in trend with regards to the incidence-density of clinical VRE isolates at Hospital 5 (coefficient: 0.004, 95%CI: -0.030 to 0.037; $P = 0.737$; $R^2 = 0.67$) and 6 (coefficient: 0.070, 95%CI: -0.066 to 0.206; $P = 0.158$; $R^2 = 0.71$). There was also no change in the trend of the incidence-density of clinical VRE isolates when data from all hospitals were combined (coefficient 0.007; 95%CI: -0.022 to 0.035; $P = 0.417$; $R^2 = 0.34$). For Hospitals 2 (coefficient -0.038; 95%CI: -0.074 to -0.001; $P = 0.047$; $R^2 = 0.91$) and 7 (coefficient -0.013; 95%CI: -0.023 to -0.003; $P = 0.029$; $R^2 = 0.94$), the incidence-density of clinical VRE cases decreased significantly over the 5-year period.

Discussion

Enterococcal resistance to vancomycin and teicoplanin is mainly due to the alteration of the amino acid composition of its cell wall terminus, resulting in reduced binding affinity. This alteration is associated with a number of genetic clusters, of which *van*A, *van*B and *van*C have primarily been found in clinical enterococcal isolates to date. The former 2 clusters are encoded on mobile genetic elements that can be transferred between *Enterococcus spp*. and even unrelated organisms such as *Staphylococcus aureus*, resulting in vancomycin-resistant *S. aureus*.[8,9] A small (<2%) proportion of VRE in this study comprised of non-*E. faecium*/non-*E. faecalis* isolates. These were labeled as VRE if the contributing laboratory had reported them as being resistant to vancomycin. Although they are grouped together with vancomycin-resistant *E. faecium* and *E. faecalis* as VRE collectively, the implications of isolating these other *Enterococcus* spp. (including *E. casseliflavus, E. durans, E. gallinarum*, etc.) differ significantly. It is important to note that they are less pathogenic and their vancomycin resistance determinants (generally *van*C) are almost invariably chromosomally mediated and therefore far less transmissible. They are not thought to pose a significant public health threat, and strict infection control measures such as patient isolation are not required if these organisms are cultured.[10]

Our surveillance showed that the overall incidence of VRE in Singapore is stable, albeit with small outbreaks identified occasionally in tertiary hospitals. As each outbreak generally resulted in increased active screening of contacts and high-risk patient populations, there was a

marked increase in the number of VRE screening isolates uncovered during those periods, along with a corresponding increase in the incidence-density of all VRE cases. However, the actual number of clinical isolates—and by extension, infections—did not increase significantly despite these outbreaks, and the prevalence of VRE clinical isolation/infections locally remains comparatively lower than in many hospitals in developed countries such as the US.[11] No formal molecular typing was done except in Hospital 5, where the vast majority of VRE isolates were *van*B-carrying *E. faecium* with identical variable-number tandem repeat profiles compared to the major outbreak VRE clones in 2005 (data not shown).[12] Given these results, it is plausible that the outbreak strains from 2005 have become entrenched in Singaporean hospitals, maintaining a low level of endemicity interspersed with occasional outbreaks.

It is less clear why local VRE clinical isolation rates have remained low compared to many western hospitals, and this is despite much higher prevalence of other antimicrobial-resistant pathogens in Singaporean hospitals.[13] Perhaps the substantial effort invested by Hospital 6 in controlling the VRE outbreak in 2005,[6] followed by enhanced measures aimed at controlling methicillin-resistant *S. aureus* in most local hospitals,[14] have prevented VRE from reaching an epidemic tipping point to date. Two of the 6 hospitals (Hospitals 1 and 3—Hospital 4 was excluded as it had been in operation for less than a year) have remained free of outbreak strains of VRE over this period. The reasons for this success are unknown. It is plausible that the different patient population (paediatrics and obstetrics/gynaecology services) of Hospital 3 prevented significant cross-transmission between this and other hospitals, whereas Hospital 1—the smallest acute care public sector hospital in Singapore at 400 beds—may have lacked the critical mass of susceptible patients for clinical VRE infections to occur. Nonetheless, it would be premature to conclude that this state of affairs will remain unchanged and heightened compliance to infection control practices as well as continued surveillance is necessary in order to forestall future outbreaks.

There are several limitations to this form of surveillance. As only microbiology laboratory reports were reviewed, it was not possible to glean the impact of infection control or antimicrobial utilisation on VRE rates. Secondly, beyond isolates from blood cultures, it was impossible to differentiate colonisation from infection in the non-screening isolates, as the majority of these VRE were obtained from non-sterile sites such as urine samples. The lack of molecular typing for the vast majority of these isolates also precluded confirmation of the presence of entrenched circulating VRE strains except at one hospital.

Conclusion

This is the first comprehensive national survey on VRE since the last major published outbreak in 2005. The prevalence of this organism remains low in Singaporean public sector hospitals. However, the presence of at least 2 small outbreaks in separate hospitals over the past 5 years indicates the need for continued vigilance in order to prevent further dissemination and corresponding increases in VRE prevalence locally.

Acknowledgements

We thank our many dedicated colleagues who have helped with data collection and technical issues, in particular Roland Jureen, Chiew Yoke Fong, Mee-Lee Tan, Winnie Lee, David Lye, Wan-Peng Lim, Hong-Yin Liew, Fiona Ooi, Chay-Leng Yeo, Serene Yeow, and Siok-Ying Lee.

This work was funded by the following grants: SingHealth Foundation Grant 2006, Ministry of Health Healthcare Quality Improvement Fund 2006, as well as education grants from Pfizer Singapore, Janssen-Cilag, Merck Sharpe & Dohme, and AstraZeneca.

L.Y.H. has received research funding and speaker's honoraria from Pfizer, AstraZeneca, Janssen & Cilag, and Merck, Sharpe & Dohme. Other authors have no conflicts of interest to declare.

REFERENCES

1. Uttley AH, Collins CH, Naidoo J, George RC. Vancomycin-resistant enterococci. Lancet 1988;1:57-8.
2. Leclercq R, Derlot E, Duval J, Courvalin P. Plasmid-mediated resistance to vancomycin and teicoplanin in Enterococcus faecium. N Engl J Med 1988;319:157-61.
3. Cetinkaya Y, Falk P, Mayhall CG. Vancomycin-resistant enterococci. Clin Microbiol Rev 2000;13:686-707.
4. Infectious Diseases Society of America. Bad bugs no drugs: as antibiotic discovery stagnates, a public health crisis brews. July 2004. Available at: http://idsociety.org/10x20.htm. Accessed 20 March 2011.
5. Chlebicki MP, Ling ML, Koh TH, Hsu LY, Tan BH, How KB, et al. First outbreak of colonization and infection with vancomycin-resistant Enterococcus faecium in a tertiary care hospital in Singapore. Infect Control Hosp Epidemiol 2006;27:991-3.
6. Kurup A, Chlebicki MP, Ling ML, Koh TH, Tan KY, Lee LC, et al. Control of a hospital-wide vancomycin-resistant enterococci outbreak. Am J Infect Control 2008;36:206-11.
7. Clinical and Laboratory Standards Institute. Analysis and presentation of cumulative antimicrobial susceptibility test data; approved guideline, 2nd ed. Document M39-A2, vol. 25, no. 28. 2006. Clinical and Laboratory Standards Institute, Wayne, PA.
8. Wood AJ. Vancomycin resistant enterococcal infections. N Engl J Med 2000;342:710-21.
9. Perichon B, Courvalin P. VanA-type vancomycin-resistant Staphylococcus aureus. Antimicrob Agents Chemother 2009;53:4580-7.

10. Tschudin Sutter S, Frei R, Dangel M, Gratwohl A, Bonten M, Widmer AF. Not all patients with vancomycin-resistant enterococci need to be isolated. Clin Infect Dis 2010;51:678-83.
11. Ramsey AM, Zilberberg MD. Secular trends of hospitalization with vancomycin-resistant enterococcus infection in the United States, 2000-2006. Infect Control Hosp Epidemiol 2009;30:184-6.
12. Koh TH, Low BS, Leo N, Hsu LY, Lin RT, Krishnan P, et al. Molecular epidemiology of vancomycin-resistant enterococci in Singapore. Pathology 2009;41:676-80.
13. Hsu LY, Tan TY, Jureen R, Koh TH, Krishnan P, Tzer-Pin Lin R, et al. Antimicrobial drug resistance in Singapore hospitals. Emerg Infect Dis 2007;13:1944-7.
14. Pereira LA, Fisher DA. Methicillin-resistant Staphylococcus aureus control in Singapore: moving forward. Ann Acad Med Singapore 2008;37:891-6.