Clinical Prevalence, Antimicrobial Susceptibility, and Geographic Resistance Patterns of Enterococci: Results from the SENTRY Antimicrobial Surveillance Program, 1997–1999

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As part of the SENTRY Antimicrobial Resistance Surveillance Program, a total of 4998 strains of enterococci isolated from 1997 to 1999 were processed. The occurrence of enterococcal infections by species and site of infection was analyzed, as were the occurrence of vancomycin-resistant enterococci (VRE) and their resistance phenotypes and genotypes. Trends in antimicrobial susceptibility to a variety of agents (including experimental compounds) were also reported. Enterococci accounted for >9% of isolates from all bloodstream infections (BSIs) in North America. Ampicillin was active against strains from Latin America and Europe but not against those from the United States and Canada. US isolates were considerably more resistant to vancomycin (17% resistant strains in 1999) than were those from patients in the rest of the world. The highest proportion of VRE was observed among BSI isolates (81.7%). Quinupristin-dalfopristin, chloramphenicol, and doxycycline were the most active agents tested against VRE. The results of this study confirm the worldwide trend in increasing occurrence of enterococci and the emerging pattern of antimicrobial resistance among such isolates.

During the past decade, enterococci have emerged as important causes of nosocomial and community-acquired infections. They were reported as the second most common cause of nosocomial infections in the United States [1]. The most frequent infections caused by enterococci are urinary tract infections. The second most frequent enterococcal infections generally have been intra-abdominal and pelvic abscesses or post-surgery wound infections. In these settings, enterococci are usually part of a mixed flora commonly found in the gastrointestinal tract, and it remains difficult to differentiate colonization from true infection. Interactions among various bacteria have been demonstrated, and studies suggest that enterococci can act synergistically with other bacteria to enhance infection. However, the role played by enterococci in these cases has not been fully defined [2], since infections of surgical wounds and of the urinary tract often resolve without specific therapy [3].

The third most frequent infection caused by these organisms is bacteremia. Other infections caused with lower frequency are CNS and neonatal infections. Enterococci rarely cause respiratory tract infections, osteomyelitis, or cellulitis [4]. Enterococci have been documented to be the third most prevalent pathogens in nosocomial bloodstream infections (BSIs) in the United States and are associated with 5%–15% of cases of bacterial endocarditis [5]. In 1996, data from the Surveillance and Control of Pathogens of Epidemiologic Importance (SCOPE) program monitoring nosocomial BSIs [6] revealed that enterococci accounted for 11.7% of all isolates, an increase in incidence of 3% over that

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reported in 1993 by the National Nosocomial Infection Surveillance (NNIS) system of the Centers for Disease Control and Prevention (CDC) [7]. The SENTRY Antimicrobial Surveillance Program carried out in 1997 in the United States, Canada, and Latin America [8] showed that enterococci were the fourth most common cause of nosocomial BSIs in participating hospitals in North America, accounting for 9.1%–9.6% of BSIs. In contrast, these organisms were isolated from only 2.9% of BSIs in Latin America and were ranked ninth overall among etiologic agents [8].

Enterococci are intrinsically resistant to many antimicrobial agents, including aminoglycosides, clindamycin, the antistaphylococcal penicillins (oxacillin, methicillin, and nafcillin), the cephalosporins, and most fluoroquinolones. Furthermore, the ability of enterococci to develop or acquire resistance to other agents is well recognized. This resistance can take many forms: high-level aminoglycoside resistance (MICs >500 μg/mL or >1000 μg/mL), β-lactamase production, high-level penicillin resistance, and resistance to chloramphenicol, tetracycline, and glycopeptides [9].

Resistance to vancomycin has been classified into 4 principal phenotypes [10–12]. The so-called VanA resistance phenotype, which is the most frequently encountered, is inducible by both vancomycin and teicoplanin and confers high-level resistance to vancomycin and teicoplanin. VanA resistance is usually plasmid borne and has genes carried on a conjugative transposon (Tn1546). The VanB phenotype shows variable levels of vancomycin resistance but remains susceptible to teicoplanin and is inducible by vancomycin alone. The genes encoding the VanB resistance phenotype are more commonly chromosomal but can also be transferred by conjugation.

Both VanA and VanB resistance phenotypes are found most frequently in *Enterococcus faecium* and *Enterococcus faecalis*. The VanC resistance phenotype is characterized by constitutive low-level resistance to vancomycin and susceptibility to teicoplanin and is routinely found in strains of *Enterococcus gallinarum* and *Enterococcus casseliflavus*. It is caused by intrinsic chromosomally encoded genes. The VanD phenotype has been noted by Perichon et al. [13] in a single strain of *E. faecium* with constitutive resistance to vancomycin and to low levels of teicoplanin. Recently, a new type of acquired glycopeptide resistance, VanE, has been described [14]; it was noted in an *E. faecalis* strain, which was resistant to low levels of vancomycin (MIC, 16 μg/mL) and was susceptible to teicoplanin.

Enterococci are components of the normal gastrointestinal tract flora of humans and animals. The nosocomial transmission of vancomycin-resistant enterococci (VRE) may occur directly between patients or indirectly via the hands of hospital staff or environmental contamination. Risk factors for acquiring infection with VRE include prolonged hospital stay, severe underlying disease, intensive care unit (ICU) stay, proximity to another patient with VRE, and treatment with multiple antimicrobial drugs such as vancomycin, third-generation cephalosporins, and some antianaerobic drugs (metronidazole, clindamycin, imipenem) [15–17].

The emergence and spread of multidrug-resistant enterococci have been seen in many countries in the past 10 years [18–22]. VRE were first reported in Europe, in 1988 [23]; they have since been identified with increasing frequency in many nations [7, 15, 24–27]. The overall prevalence of vancomycin resistance among enterococcal isolates from hospitals in 6 geographic regions of the United States collected from July 1988 through April 1989 was very low (0.3%) [28]. A national surveillance study conducted to evaluate antimicrobial resistance among enterococci in US hospitals in 1992 revealed that 4.4% of isolates were resistant to vancomycin [29]. The percentage of vancomycin-resistant nosocomial enterococcal infections reported to the CDC-NNIS system increased from 0.3% in 1989 to 7.9% in 1993 [1]. This increase was higher among patients in ICUs, rising from 0.4% to 13.6% in the same time interval. In some hospitals, VRE became established as an endemic nosocomial pathogen [30].

Results from the 1997 SENTRY program revealed that 14.1% of enterococcal BSIs in the United States were attributable to VRE [8]. In the SCOPE project on nosocomial BSIs in 49 US hospitals over the period 1995–1998, 17.7% of enterococcal isolates displayed resistance to vancomycin. The proportion of resistance to vancomycin was 16-fold higher (50.5%) among *E. faecium* isolates than among *E. faecalis* isolates (3.1%) [31].

Surveillance programs such as the CDC-NNIS, SCOPE, and Intensive Care Antimicrobial Resistance Epidemiology (ICARE) projects have provided data that were limited by their focus on nosocomial infections (NNIS and SCOPE) and/or by the absence of validated identification or antimicrobial susceptibility testing in a central or reference laboratory (NNIS and ICARE). The SENTRY program was designed to monitor the spectrum of microbial pathogens concurrent with antimicrobial resistance trends for both nosocomial and community-acquired infections on a global scale [32].

Previous publications have described the results of various aspects of the SENTRY program [8, 31, 33–35]. The present report will focus on enterococcal isolates recovered at medical centers in 5 geographic regions (the United States, Canada, Latin America, Europe, and the Asia-Pacific) from 1997 to 1999. Three regional monitoring laboratories confirmed the organism identifications and performed reference-quality antimicrobial susceptibility testing. This report discusses the occurrence of enterococcal infections by species and site of infection, as well as the occurrence of VRE isolates and their resistance characterization. Trends in susceptibility to a wide variety of antimicrobials (including experimental agents) in each region and demographic profiles are also reported.

MATERIALS AND METHODS

Study design. The SENTRY Antimicrobial Surveillance Program was initiated in early 1997 to investigate the longitudinal trends in antimicrobial resistance and the frequency of pathogen occurrence. A total of 5 major objectives address the most common types of infection, in a prevalence-style format: objective A, BSIs; objective B, community-acquired respiratory tract infections caused by *Streptococcus pneumoniae, Haemophilus influenzae,* and *Moraxella catarrhalis;* objective C, pneumonias in hospitalized patients; objective D, skin and soft-tissue or wound infections; and objective E, urinary tract infections. Consecutive isolates (540 strains/year for all objectives per laboratory) were forwarded to the regional monitors for reference-quality antimicrobial susceptibility testing and confirmation of organism identification. Nearly 100,000 bacteria have been processed, the vast majority within the objective (A–E) protocols (>70,000; table 1). A total of 4998 strains of enterococci were processed, each accompanied by key demographic parameters.

Participants and monitors. Three reference laboratories acted as monitor sites during the 1997–1999 interval: the University of Iowa College of Medicine, in Iowa City, Iowa (North and Latin America regions for 1997–1999 and Europe region for 1999); Utrecht University, in Utrecht, The Netherlands (Europe region for 1997–1998); and the Women's and Children's Hospital, in Adelaide, Australia (Asia-Pacific region for 1998–1999). Common reagents and data processing systems were used.

The participating sites varied slightly in number by year and included the following: 5–8 sites in Canada (North American region); 26–28 sites in the United States (United States/North American region), 10 sites in the Latin American region; 12–23 sites in Europe, Israel, and Turkey (Europe region); and 17 sites in the Asia-Pacific region (also includes 1 site in South Africa). The number of participants worldwide ranged from 66 laboratories in 1997 to 81 laboratories in 1998.

Antimicrobial agents. Representatives from all clinically important antimicrobial classes have been tested (ampicillin, penicillin, erythromycin, vancomycin, teicoplanin, chloramphenicol, doxycycline, gentamicin, streptomycin, ciprofloxacin, nitrofurantoin, and trimethoprim-sulfamethoxazole [TMP-SMZ]), as well as investigational compounds such as linezolid (U-100766), evernimicin (SCH 27899), quinupristin-dalfopristin (Synercid; Aventis), and gatifloxacin. Antimicrobials were obtained from their US manufacturer or representative and were dispensed into validated dry-form broth microdilution trays (MicroScan; TREK/Sensititre). Each lot of trays was shared among all monitor sites, and quality-control results were satisfactory in all cases.

Antimicrobial susceptibility testing. All susceptibility testing was performed with the above-cited reagents in Mueller-Hinton broth and according to the methods recommended in documents by the National Committee for Clinical Laboratory Standards (NCCLS) [36, 37]. All quality-control determinations (daily) were within published guidelines [38].

Identifications of the species were initially determined at the participating sites and were confirmed by means of the Vitek System (bioMérieux Vitek) [39, 40] or conventional test procedures [41–43]. Where discords were suspected, a PCR species-identification method was employed [44]. The PCR procedure was adapted to a robot to minimize test-to-test variability and possible contamination from human technical staff [45].

The determination of various *van* genotypes was also accomplished by PCR [44] with the use of methods and primers described elsewhere [6].

Table 1. Variations in the occurrence of enterococcal infections in hospitals contributing isolates to the SENTRY Antimicrobial Surveillance Program (1997–1999).

	No. of isolates per site of infection, % enterococci (range[a])			
Country or region	Blood	Respiratory	Wound	Urine
Canada	3840	1659	633	630
	9.2 (8.8–9.9)	0.3 (0.0–0.6)	5.1 (4.1–6.5)	16.8 (15.4–17.8)
Europe	10,815	2572	2305	2135
	6.8 (6.7–7.3)	1.7 (1.3–2.9)	6.7 (6.7–6.8)	11.7 (11.4–12.0)
Latin America	5295	1914	1353	1430
	3.0 (2.7–3.3)	2.0 (1.8—2.3)	8.0 (6.6–9.7)	4.2 (3.6–4.9)
United States	17,399	6711	2191	2569
	9.9 (9.6–10.6)	0.9 (0.7–1.2)	9.1 (8.8–9.4)	12.5 (12.4–12.6)
Asia-Pacific[b]	3162	1704	791	959
	4.6 (3.9–5.3)	2.6 (2.2 –3.4)	4.8 (1.0–6.1)	10.7 (10.1–11.1)

NOTE. A total of 70,067 strains (4998 enterococci) were analyzed over the 3 study years.

[a] Range indicates occurrence rates over the 3 years; minimal variations were observed.

[b] 1998 statistics only.

RESULTS

Nationwide surveillance programs, such as the SCOPE program [6], have provided data on nosocomial enterococcal BSIs, and the NNIS system [1, 7] also provides national data on various types of nosocomial infections. The SENTRY program has shown occurrence rates for 4998 enterococci strains by site of infection (blood, respiratory tract, wound, and urinary tract) and is the only surveillance system that yields data on a broad geographic scale including the 5 geographic regions. During the study period (1997–1999), a total of 70,067 strains were analyzed by SENTRY participants (6762 from Canada; 17,827 from Europe; 9992 from Latin America; 28,870 from the United States; and 6616 from the Asia-Pacific region).

Table 1 shows the occurrence rates of enterococcal infections by site of infection within each geographic region. Enterococci accounted for >9% of BSI isolates in Canada and the United States. The lowest rates of enterococcal infections in bloodstream and urinary tract sites were discovered in Latin America (3.0% and 4.2%, respectively). The highest detected rate of enterococcal urinary tract infection was in Canada (16.8%), followed by the United States (12.5%) and Europe (11.7%).

The frequency of reported isolations of *Enterococcus* species, by geographic region, is listed in table 2. Of the 8 reported species, *E. faecalis* was the most prevalent, representing from 57.2% of enterococci isolated in Canada to 76.8% in Latin America. *E. faecium,* the species in which vancomycin resistance is most prevalent, was the second most commonly identified species in all geographic regions. The proportion of isolates of species intrinsically resistant to vancomycin *(E. casseliflavus* and *E. gallinarum)* continued to be low and varied with geographic region (range, 0.6%–2.2%). Identification to the species level was not performed for 3.6%–21.2% of the isolates reported by the laboratories of each of the 5 regions.

Trends in antimicrobial susceptibility of the enterococcal strains tested are shown in table 3. Ampicillin was most active against strains from Latin America and Europe (84%–99% susceptible) but not against strains from the United States and Canada (76% susceptible in 1999), where *E. faecium* was most prevalent (19.3%–20% of enterococcal isolates). US isolates were considerably more resistant to vancomycin than were those from the rest of the world. In Latin America all isolates tested in 1997 were susceptible to vancomycin, although the rate of resistance to this glycopeptide was 1% in 1998, increasing to 2% a year later. In the remaining geographic regions, the rates of vancomycin resistance were very low (1%–3%) and did not change appreciably during the reported study period.

Teicoplanin was more active than vancomycin against the US isolates, and this finding correlates with the occurrence of VanB strains of vancomycin resistance. Geographic variations in chloramphenicol resistance were also observed, and susceptibility rates were 10%–20% lower in the Latin America, Europe, and Asia-Pacific regions than among strains in Canada and the United States. Doxycycline was active against one-half of strains tested. The highest doxycycline resistance rates were detected among the Asia-Pacific strains. As high-level resistance was lower for gentamicin than for streptomycin, gentamicin was the most usable drug overall for coadministration, especially in Latin America, where isolates showed the lowest resistance to aminoglycosides.

The 2 fluoroquinolones tested, ciprofloxacin and gatifloxacin, displayed reduced activity against the nearly 5000 strains processed in this study. The resistance rates to gatifloxacin were generally lower than those of ciprofloxacin. Overall, the isolates from Latin America tended to be more susceptible to gatifloxacin than did the European, Asia-Pacific, or North American isolates (rank order: Latin America [most susceptible] > Asia-Pacific = Europe > North America [least susceptible]).

Nitrofurantoin was active against the majority of strains tested and remains an acceptable candidate for treatment of enterococcal urinary tract infections. TMP-SMZ showed poor

Table 2. Incidence of *Enterococcus* species isolations from all monitored infections in 5 geographic regions in the SENTRY Antimicrobial Surveillance Program (1997–1999).

	Incidence (%) among enterococci isolates tested, by region				
Enterococcus species	Canada (n = 509)	United States (n = 2400)	Latin America (n = 367)	Europe (n = 1201)	Asia-Pacific (n = 331)
E. avium	0.6	0.8	1.6	1.0	3.3
E. casseliflavus	0.4	0.3	0.3	0.8	1.2
E. durans	0.2	0.1	0.3	1.6	0.0
E. faecalis	57.2	60.0	76.8	67.9	75.8
E. faecium	19.3	20.0	4.6	14.7	14.8
E. flavescens	0.0	<0.1	0.0	0.0	0.0
E. gallinarum	1.0	0.7	0.3	0.4	1.2
E. raffinosus	0.2	0.2	0.0	<0.1	0.0
Undetermined	21.2	18.0	16.1	13.5	3.6

Table 3. Trends in antimicrobial susceptibility of all tested enterococci (nearly 5000 strains) in each monitored region for 1997, 1998, and 1999 (SENTRY Antimicrobial Surveillance Program).

Antimicrobial agent	Isolates (%) susceptible[a]													
	Canada			United States			Latin America			Europe			Asia-Pacific	
	1997	1998	1999	1997	1998	1999	1997	1998	1999	1997	1998	1999	1998	1999
Ampicillin	82	78	76	79	78	76	97	93	99	88	84	88	86	80
Vancomycin	99	98	100	86	86	83	100	99	98	97	97	99	98	99
Teicoplanin	99	100	100	90	90	86	100	99	98	97	97	99	>99	100
Chloramphenicol	77	69	86	81	76	88	66	68	73	71	65	68	67	74
Doxycycline	59	53	50	53	51	54	54	49	39	58	44	42	30	39
Gentamicin-HL[b]	60	60	65	71	72	69	68	86	84	71	72	74	64	70
Streptomycin-HL[b]	60	55	64	61	60	60	64	70	65	61	53	54	63	72
Ciprofloxacin	40	25	45	40	36	42	35	47	43	58	58	49	42	50
Gatifloxacin	55	47	48	57	57	51	62	81	89	69	68	74	70	72
Nitrofurantoin	78	82	82	79	86	81	85	94	92	92	88	88	90	87
TMP-SMZ	54	51	67	62	64	63	69	81	88	73	78	80	83	79

NOTE. TMP-SMZ, trimethoprim-sulfamethoxazole.

[a] Based on National Committee for Clinical Laboratory Standards (2000) interpretive criteria for susceptibility.

[b] HL, high-level resistance to aminoglycosides (susceptibility rates are listed).

activity against Canadian strains isolated in the first and second years of the study (54% and 51% susceptible, respectively). It is notable that among US strains, ampicillin and vancomycin resistance continue to increase at alarming rates and are currently at 17% and 24%, respectively.

The incidence of vancomycin-resistant isolates ($n = 409$), listed by resistance phenotype and by the body site of infection, is shown in table 4. The highest proportion of VRE was seen in BSIs (81.7% of isolates), and lower proportions were observed in urinary tract (8.8%), wound (7.6%), and respiratory tract infections (1.9%). The number of isolations in the United States (344 strains from ~30 medical centers) was clearly greater than in Canada (7 strains from 8 sites) and elsewhere in the monitored regions. Overall, 73.8% of VRE exhibited the VanA phenotype, 21.3% the VanB phenotype, and 4.9% the VanC phenotype. The highest proportion of VanA phenotype, however, was observed among European strains (90%).

Figure 1 shows the number of VRE isolated in North America during 1997–1999 in 5 US geographic regions and Canada. The north-central and northeast regions yielded the most VRE, with the greatest occurrence during 1998. A recent decline in VRE (in 1999) occurred in all US regions and Canada. This decrease could be due in part to the implementation of the recommendations established by the Hospital Infection Control Practices Advisory Committee (HICPAC) in collaboration with the CDC [46]. These include the following key elements: prudent use of vancomycin, education of hospital staff, early detection and prompt reporting of resistant strains by the microbiology laboratory, and the implementation of effective infection control measures [47]. Educational programs to improve the use of antimicrobials as well as continued surveillance of antimicrobial susceptibility patterns are needed in hospitals in order to maintain optimal infection control policies and procedures and to guide interventions. Important efforts have been made to emphasize the need for prevention and control measures. Greater physician awareness of all prevention policies and measures to control the spread of VRE could also have contributed to the recent decline in prevalence of VRE observed in all US regions and Canada.

Additional SENTRY data about VRE in the United States and elsewhere are as follows: (1) 33 participating sites in the United States reported strains over 3 years, and the number of isolates reported ranged from 1 (4 sites) to 40; (2) 6 medical centers had ⩾20 isolates during the study period, and 4 of these 6 hospitals had fewer VRE isolates in 1999; (3) VRE in Canada came from 4 of the 8 medical centers monitored during 1997–1999; (4) only 3 sites in Latin America (in 3 different nations) isolated VRE, and all strains appeared in 1998 and 1999; and (5) VRE in Europe were isolated at 7 hospitals (in Greece, Israel, Spain, Portugal, and the United Kingdom, as well as 2 in Italy). The highest number of isolates (19) was in Portugal.

In all, 50 participating medical centers have reported VRE to the SENTRY program. Epidemiological or endemic VRE clusters could be suspected in 6 US medical centers that reported ⩾20 VRE isolates during the study period, as well as the hospital in Portugal. Further studies with use of molecular typing methods would be necessary to assess the suspected clusters; all cited locations had clonal spread proven by applied ribotyping and pulsed-field gel electrophoresis (University of Iowa).

Demographic data on VRE were available from all regions

Table 4. Incidence (number) of vancomycin-resistant enterococci, by type of resistance phenotype and body site of isolation (SENTRY Antimicrobial Surveillance Program, 1997–1999).

Country or region, year (no. of isolates tested)	Site of infection				Vancomycin-resistant phenotype		
	Blood	Respiratory	Wound	Urine	VanA	VanB	VanC
Canada							
1997 (n = 3)	2	0	0	1	2	0	1
1998 (n = 4)	4	0	0	0	0	3	1
1999 (n = 0)	0	0	0	0	0	0	0
United States							
1997 (n = 129)	105	2	11	11	86	39	4
1998 (n = 115)	94	1	9	11	87	21	7
1999 (n = 110)	95	4	4	7	88	20	2
Latin America							
1997 n = 0)	0	0	0	0	0	0	0
1998 (n = 1)	0	0	0	1	0	0	1
1999 (n = 2)	2	0	0	0	2	0	0
Europe							
1997 (n = 18)	13	0	3	2	17	1	0
1998 (n = 20)	15	0	3	2	19	1	0
1999 (n = 2)	2	0	0	0	0	2	0
Asia-Pacific[a]							
1998 (n = 3)	0	1	1	1	1	0	2
1998 (n = 2)	2	0	0	0	0	0	2

NOTE. Data are for 409 isolates from 5 regions.

[a] Data from 1998 only.

except Europe (40 strains; most [75%] were from BSIs) and Asia-Pacific (only 5 strains). Among the rare VRE isolates from Canada and Latin America, 71% and 67%, respectively, were nosocomial. The following demographics were documented from the United States: (1) Of the 354 VRE isolates, 81.6% were from BSIs. (2) The male-to-female ratio varied by year (the percentages of males were 55.8 [1997], 47.8 [1998], and 61.8% [1999]). (3) The age range was wide (1 month to 96 years), but the average age each year varied only between 57 and 58 years. (4) Isolates from ICU patients accounted for 61% of strains in 1997 but significantly fewer (36%–44%) in 1998–1999. (5) The nosocomial acquisition rate was 73%–76%. (6) When the patient ward or service was known, the predominant areas were internal medicine (40%), hematology-oncology (17%), and gastrointestinal-surgery (16%).

As resistance to vancomycin is usually accompanied by multiple resistance to other antimicrobial agents such as macrolides, tetracyclines, and the newer fluoroquinolones, the activity of alternative therapeutic agents for VRE infections was evaluated. Table 5 lists the median MIC (or MIC_{50}, in μg/mL) and percentage of isolates susceptible for 12 antimicrobial agents tested against all VRE in the SENTRY program, by geographic region. The most active agent was quinupristin-dalfopristin (71%–82% susceptible, among US and Canada isolates), followed by chloramphenicol (33%–90% susceptible) and doxycycline, which inhibited 40%–65% of VRE strains tested from all but 1 geographic region (Latin America). The VRE isolates from Latin America were uniformly more susceptible to ampicillin and penicillin. A large proportion of the isolates (80%–100%) were resistant to erythromycin. Although gatifloxacin was superior to ciprofloxacin, fewer than one-third of strains were susceptible.

Potential synergy with aminoglycosides was highest with gentamicin (33%–57%), but this rate was lower than that observed among all enterococci tested (table 3). High-level resistance to gentamicin was more common among Latin American VRE isolates (33% susceptible) than among isolates from the other 4 regions. In contrast, high-level resistance to streptomycin was more common among Canadian and US isolates (14% and 21% susceptibility, respectively). Nitrofurantoin remained active against VRE infections in the urinary tract, especially in Europe (83% of isolates susceptible). The highest susceptibility to TMP-SMZ was observed among VRE isolates from Latin America and the Asia-Pacific region (67% and 80%, respec-

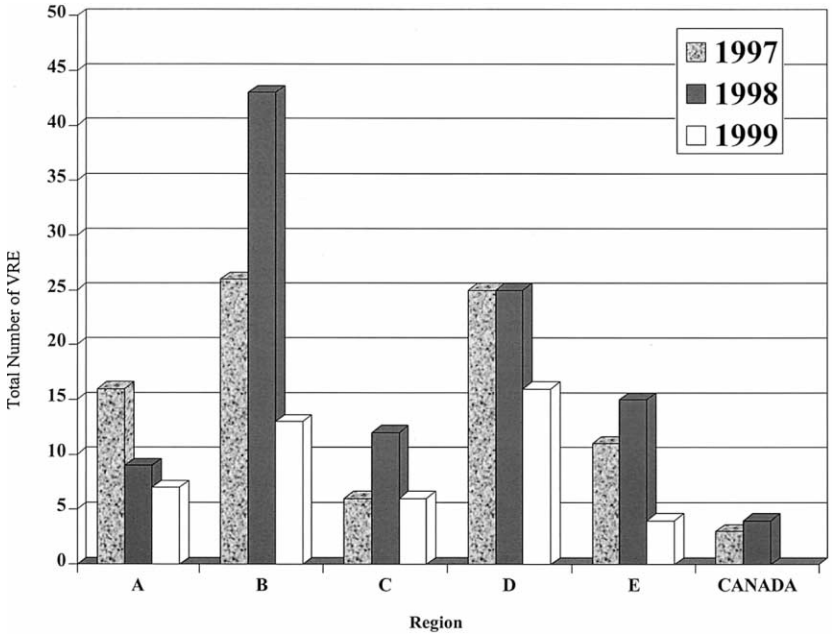

Figure 1. Isolations of vancomycin-resistant enterococci (VRE) at SENTRY program medical centers in the 5 regions of the United States (A–E) and in Canada during 1997–1999 (A, west; B, north-central; C, south-central; D, northeast; E, southeast).

tively). Linezolid and evernimicin (SCH 27899) were active against all strains (data not shown), as previous studies have demonstrated [48, 49].

There are few alternatives for the treatment of infections caused by multidrug-resistant *E. faecium*. Several studies have documented the excellent in vitro activity of quinupristin-dalfopristin against multidrug-resistant gram-positive pathogens, such as methicillin-resistant *S. aureus*, vancomycin-resistant *E. faecium*, glycopeptide-resistant coagulase-negative staphylococci, and pneumococci resistant to penicillin and macrolides [8, 50, 51]. In a previous study [51], the streptogramin combination was tested against nearly 30,000 clinical strains of gram-positive organisms from a total of 200 medical center laboratories in the United States and Canada. Quinupristin-dalfopristin showed excellent in vitro activity against nearly all strains tested. The investigators demonstrated that enterococcal species misidentification was common by retesting all available strains that were resistant to quinupristin-dalfopristin. The error rate among all retested *E. faecium* strains was 94.7%, and only 0.2% of all referred enterococcal isolates were authenticated as resistant (MIC, $\geq$4 μg/mL) to quinupristin-dalfopristin.

Although identification error [39] may contribute to false-positive values for resistance to quinupristin-dalfopristin for *E. faecium* [51], true resistance to this new streptogramin was observed in the SENTRY program (table 6). A total of 134 *E. faecium* strains were confirmed as nonsusceptible to quinupristin-dalfopristin (MIC, $\geq$2 μg/mL; 49 from North America, 68 from Europe, 7 from Latin America, and 10 from Asia-Pacific). However, the majority of them showed intermediate susceptibility to this agent (MIC, 2 μg/mL), and only 15 strains had high-level resistance (MICs, $\geq$8 μg/mL). Not all quinupristin-dalfopristin-nonsusceptible *E. faecium* isolates were resistant to vancomycin; in fact, for 74.4% (United States) to 100.0% (Canada, Latin America, and the Asia-Pacific region) of isolates, the vancomycin MICs were $\leq$4 μg/mL. Ampicillin also inhibited 39.7%–80% of strains, depending on the geographic region.

As can be seen by comparing the results in table 6 with those in table 5, quinupristin-dalfopristin-nonsusceptible *E. faecium* strains were more likely to be free of high-level aminoglycoside resistance than were all VRE isolates tested. The emergence of increased resistance to quinupristin-dalfopristin observed during therapy for *E. faecium* bacteremia [52] was probably related to the slow bactericidal effects of this streptogramin combination on *E. faecium* strains resistant to macrolides [53]. This fact must be considered when therapy against multiresistant *E. faecium* is to be selected.

DISCUSSION

Despite the fact that enterococci have been considered to have relatively low virulence, in the past few years these organisms, among all nosocomial pathogens, have emerged as a significant concern. VRE may cause a range of infections associated with high mortality [54]. The study by Edmond et al. [55] demonstrated the high morbidity and mortality attrib-

Table 5. Potency and spectrum of 12 selected antimicrobial agents tested against 409 vancomycin-resistant enterococcal isolates in the SENTRY Antimicrobial Surveillance Program (1997–1999).

Antimicrobial agent	MIC_{50}, μg/mL (% of tested isolates susceptible)				
	Canada (*n* = 7)	United States (*n* = 354)	Latin America (*n* = 3)	Europe (*n* = 40)	Asia-Pacific (*n* = 5)
Ampicillin	>16 (29)	>16 (12)	1 (100)	>16 (35)	1 (60)
Penicillin	>32 (29)	>32 (10)	4 (100)	>32 (35)	2 (60)
Erythromycin	>8 (14)	>8 (3)	>8 (0)	>8 (3)	>8 (20)
Quinupristin-dalfopristin	1 (71)	0.5 (82)	>8 (0)	1 (50)	2 (20)
Chloramphenicol	8 (86)	8 (90)	>16 (33)	8 (68)	8 (80)
Doxycycline	>4 (43)	4 (53)	>4 (0)	0.5 (65)	>4 (40)
Ciprofloxacin	>2 (0)	>2 (3)	>2 (0)	>2 (10)	>2 (0)
Gatifloxacin	>4 (14)	>4 (6)	>4 (33)	>4 (23)	0.5 (60)
Gentamicin-HL[a]	⩽500 (57)	>1000 (40)	>1000 (33)	⩽500 (50)	⩽500 (60)
Streptomycin-HL[a]	>2000 (14)	>2000 (21)	>2000 (33)	2000 (48)	>2000 (40)
Nitrofurantoin	64 (29)	64 (40)	⩽32 (67)	⩽32 (83)	⩽32 (60)
TMP-SMZ	>1 (14)	>1 (28)	⩽0.5 (67)	>1 (30)	⩽0.5 (80)

NOTE. TMP-SMZ, trimethoprim-sulfamethoxazole.

[a] HL, high-level resistance to aminoglycosides (susceptibility rates are listed), indicating potential synergy.

utable to VRE bacteremia. VRE are often concomitantly resistant to multiple antimicrobial classes. Increasing high-level resistance to penicillin, ampicillin, and aminoglycosides has been documented in recent years, particularly in strains of vancomycin-resistant *E. faecium* [29].

The emergence of VRE is a cause for concern because of the limited therapeutic options for treating serious infections and because of their potential to transfer vancomycin-resistance genes to other organisms, such as methicillin-resistant *Staphylococcus aureus.* Conjugative in vitro transfers of resistance genes to *S. aureus, Listeria monocytogenes,* and group A and viridans streptococci have been reported [56, 57]. VanA genes were found in *Oerskovia turbata* and *Arcanobacterium haemolyticum* isolated from stool specimens [58], and Poyart et al. [59] demonstrated the presence of a VanB-related gene in a *Streptococcus bovis* clinical isolate.

Clinical isolates of *S. aureus* with reduced susceptibility to vancomycin (MICs, 8 μg/mL) were recovered in Japan [60] and the United States [61]. In 1988, Schwalbe et al. [62] described the emergence of coagulase-negative staphylococci that displayed a stepwise increase in vancomycin resistance in a patient undergoing long-term therapy. These findings illustrate the need for antimicrobial agents with potent activity against these multiresistant organisms as well as the importance of implementation of measures to detect, control, and prevent the spread of vancomycin resistance [46, 63].

Enterococci are increasingly responsible for nosocomial infections, especially BSIs [6–8]. In the SENTRY program (1997–1999), enterococci accounted for >9% of all isolated organisms causing BSIs in the United States and Canada and for only 3.0% of BSIs in Latin America. These frequencies are similar to those in previous SENTRY reports [8, 32] but slightly lower than those reported following a nationwide surveillance of nosocomial BSI, in which enterococci accounted for 11% of all isolates [6, 31]. The most frequently isolated species was *E. faecalis,* followed by *E. faecium.* The frequency with which *Enterococcus* species were found in this study did not vary significantly from the frequencies in several surveillance studies over the past decade, especially among BSIs [6, 8, 29, 31, 32].

This study also documents differences in the rates of resistance to antimicrobials among enterococci from one geographic region to another. Overall, isolates from Canada and the United States were less susceptible to TMP-SMZ, gatifloxacin, and nitrofurantoin than were isolates from Latin America, Europe, and the Asia-Pacific region. Meanwhile, isolates from Latin America were more resistant to doxycycline than were isolates from the other 4 regions. Vancomycin resistance was significantly higher among US isolates than among those from the rest of the world.

There are differences between the United States and Europe in the incidence of both VRE colonization and VRE infection [64, 65]. The incidence of VRE infections is high in the United States in comparison with that in Europe, probably because the rate of use of vancomycin in United States hospitals has increased dramatically in recent years [66–68]. In contrast, VRE colonization in the community has not been widespread in the United States [69]. Several studies in European countries [70–73] revealed a high prevalence of VRE among inpatients and nonhospitalized persons, as well as in raw sewage and farm animals [69–71, 74–76]. In North America, VRE has not been as frequently recovered from animal sources [77].

These geographic differences might be due to the use of the

Table 6. Analysis of quinupristin-dalfopristin-nonsusceptible *E. faecium* strains (134 isolates).

Geographic region (no. of isolates tested)	Antimicrobial agent	No. of isolates at specified MIC, in μg/mL						Susceptible,[a] %
		0.5	1	2	4	8	⩾16	
Canada (n = 10)[b]	Quinupristin-dalfopristin	0	0	7	3	0	0	0.0
	Ampicillin	1	1	2	3	1	1	80.0
	Vancomycin	6	2	2	0	0	0	100.0
	Teicoplanin	9	1	0	0	0	0	100.0
	Gentamicin-HL	—	—	—	—	—	—	100.0
	Streptomycin-HL	—	—	—	—	—	—	60.0
Europe (n = 68)[c]	Quinupristin-dalfopristin	0	0	42	12	7	7	0.0
	Ampicillin	5	7	6	6	3	41	39.7
	Vancomycin	26	26	8	1	1	6	89.7
	Teicoplanin	56	6	1	0	0	5	92.6
	Gentamicin-HL	—	—	—	—	—	—	75.0
	Streptomycin-HL	—	—	—	—	—	—	57.4
Latin America (n = 7)[d]	Quinupristin-dalfopristin	0	0	5	2	0	0	0.0
	Ampicillin	0	1	1	2	1	2	71.4
	Vancomycin	3	2	1	1	0	0	100.0
	Teicoplanin	6	1	0	0	0	0	100.0
	Gentamicin-HL	—	—	—	—	—	—	71.4
	Streptomycin-HL	—	—	—	—	—	—	28.6
United States (n = 39)[d]	Quinupristin-dalfopristin	0	0	31	7	1	0	0.0
	Ampicillin	3	9	11	2	1	13	66.7
	Vancomycin	17	8	4	0	2	8	74.4
	Teicoplanin	26	3	1	1	0	8	79.5
	Gentamicin-HL	—	—	—	—	—	—	76.9
	Streptomycin-HL	—	—	—	—	—	—	64.1
Asia-Pacific (n = 10)[e]	Quinupristin-dalfopristin	0	0	8	2	0	0	0.0
	Ampicillin	0	1	2	0	1	6	40.0
	Vancomycin	3	6	1	0	0	0	100.0
	Teicoplanin	7	2	1	0	0	0	100.0
	Gentamicin-HL	—	—	—	—	—	—	80.0
	Streptomycin-HL	—	—	—	—	—	—	80.0

NOTE. HL, high-level resistance to aminoglycosides.

[a] Interpretations on the basis of National Committee for Clinical Laboratory Standards (1999) documents.
[b] Includes isolates from 5 medical centers (63% of sites).
[c] Includes isolates from 15 medical centers (75% of sites).
[d] Includes isolates from 15 medical centers (50% of sites).
[e] Strains from 4 different participants (27% of sites).

glycopeptide avoparcin as a growth promoter in animal feeds in some European countries, but this agent has not been approved for use in the United States. The use of antimicrobial agents as growth promoters in feed animals in some areas may have contributed to the emergence of antimicrobial resistance in humans. The study report of van den Boogaard et al. [78] cited the high prevalence of VRE in healthy persons living in areas where avoparcin is used as a feed supplement. A recent study [76] showed a significantly higher prevalence than that reported by other European and US investigators of VRE colonization of fecal samples obtained from hospitalized and healthy nonhospitalized subjects living in the same local community.

In US hospitals, many VRE outbreaks are due to intrahospital and interhospital spread of clonal strains [79]. This indicates that patient-to-patient transmission is the major factor responsible for VRE dissemination and persistence in the United States [80]. Further studies are required to clarify the role of community transmission of VRE within and between animal and various human populations [65].

Among European isolates, the vancomycin resistance rate observed in this study remains low (1%–3%). This is in ac-

cordance with a recent survey of the European VRE Study Group [81], in which resistance to vancomycin was detected in 0.06% of *E. faecalis* and in 3.8% of *E. faecium* strains. The overall prevalence of high-level resistance to gentamicin among European isolates is comparable to that reported from the SENTRY program. In contrast, of 177 *E. faecium* strains from Europe, 68 (38.4%) were resistant to quinupristin-dalfopristin; this rate is significantly higher than that (2%) reported by the European VRE Study Group [81].

Resistance to vancomycin among US enterococcal isolates ranged from 14% in 1997 to 17% in 1999. Similar rates have also been detected in a recent nosocomial-BSI surveillance study [31]. Teicoplanin resistance was less frequently observed (10%–14%), indicating the occurrence of VanB phenotypes [6]. The high-level resistance to gentamicin and streptogramin did not differ significantly from that observed in previous surveillance studies performed since 1995 [6], but it was slightly higher than that among strains isolated in 1992 (26.9% and 35.7%, respectively) [29] and in 1999 (29% and 40%, respectively). The susceptibilities of US strains isolated in 1999 to other alternative drugs such as ciprofloxacin (42%) and TMP-SMZ (63%) were higher than those observed in earlier national surveillance [29]. Among VRE isolates from the United States, the resistance rates to chloramphenicol and ciprofloxacin and the high-level resistance to gentamicin observed in this study were similar to those reported by Evans et al. [82], but the activity of doxycycline was lower (53% susceptible [82]).

Our results confirm the increasing trend toward antimicrobial resistance among enterococci over the world. Of concern are the high rates of resistance to vancomycin and to ampicillin among US isolates, which increased from 14% and 21%, respectively, in 1997 to 17% and 24% in 1999 (increases of 1% per year). Another concern is the emergence of resistance to glycopeptides in Latin America that was first detected in 1998 at a very low rate (1%) but doubled to 2% in 1999. Among other resistance trends detected in this study is the increasing frequency of ampicillin, doxycycline, and gatifloxacin resistance among Canadian isolates and of ampicillin, nitrofurantoin, and doxycycline resistance among Asia-Pacific isolates. An increasing trend in resistance to chloramphenicol among European isolates and to doxycycline among Latin American and European isolates was also observed.

Of greater concern is the multiple-drug resistance detected among VRE isolates, as noted in most published surveillance studies [6, 8, 29, 32, 79]. This includes high-level aminoglycoside and penicillin resistance, as well as resistance to ampicillin and other alternative agents. Resistance to vancomycin and other acquired or intrinsic resistances limit treatment choices [83]. Quinupristin-dalfopristin has been considered a therapeutic alternative for infections caused by multidrug-resistant *E. faecium* strains [35]. An increasing trend of resistance to quinupristin-dalfopristin has been detected by comparison of results of this study with those of previous investigations. Bonilla et al. [50] reported that all vancomycin-resistant *E. faecium* were highly susceptible to streptogramin. In a previous SENTRY surveillance report on gram-positive cocci causing BSIs in 1997 [8], there were no quinupristin-dalfopristin-resistant strains observed among isolates confirmed to be *E. faecium*. Only 0.2% of *E. faecium* isolates were validated as truly resistant to quinupristin-dalfopristin in the Synercid Microbiological Assessment of Resistance Trends study [51]. In our study, 103 (12.6%) of the *E. faecium* isolates showed intermediate susceptibility to quinupristin-dalfopristin (MICs, 2 μg/mL), and 31 (3.8%) were resistant (MICs, $\geq$4 μg/mL).

The increasing trend of antimicrobial resistance among enterococci processed in the SENTRY program, as previously reported following other surveillance trials [6, 18, 29, 32], highlights the importance of the implementation of infection control measures [46] in each medical center. The microbiology laboratory plays an important role in the detection, timely reporting, and control of VRE. It is essential for the microbiology laboratory to ensure that enterococci are rapidly and accurately identified to the species level [43]. Accurate testing of susceptibility should also be performed [84]. For early detection, laboratories should use reliable conventional, reference broth, and agar methods rather than automated or commercial systems, which have performed poorly in the past [85]. The difficulty in detecting low-level vancomycin resistance by disk diffusion has also been reported [86]. The HICPAC document [46] emphasizes the need for routine susceptibility testing of all enterococci isolated from clinical specimens.

Furthermore, in hospitals where VRE have not yet been detected, periodic culture surveys of stools or rectal swabs from patients at high risk of VRE infection or colonization are recommended. In vitro testing of enterococcal isolates should be performed against ampicillin and vancomycin and for high-level resistance to aminoglycosides (gentamicin and streptomycin), according to established NCCLS guidelines [36–38]. For isolates with high-level resistance to all of these agents, susceptibility to alternative agents such as teicoplanin, quinolones, chloramphenicol, doxycycline, quinupristin-dalfopristin, and nitrofurantoin (now linezolid) should be considered [3].

The rational use of antimicrobials is the most important means of minimizing enterococcal infections. The HICPAC document [46] has also emphasized the importance of education in controlling the spread of vancomycin resistance. Educational programs for health care workers to improve the use of antimicrobials, including vancomycin, are needed [68] and should be repeated periodically for reinforcement. Infection control measures are crucial to limit the nosocomial spread of

VRE. These include hand washing, barrier precautions, and isolation or grouping of infected or colonized patients [80].

In the past 2 decades, antimicrobial resistance has emerged as an important concern all over the world [87, 88]. This has been associated with changing patterns of pathogen occurrences [89]. Longitudinal surveillance programs in which reference susceptibility methods are used are essential, as they could guide physicians toward appropriate therapeutic antimicrobials. They could also influence the use of interventions necessary to reduce or stabilize resistance rates and minimize the emergence or spread of resistance genes [90].

Actions necessary to limit the selection of resistance were summarized in the comprehensive report by the American Society for Microbiology (ASM) Task Force on Antibiotic Resistance in 1995 [90]. The major components of the ASM task force recommendations are (1) surveillance networks on a local, national, and global scale in order to recognize emerging resistance and to allow appropriate data-based interventions; (2) education of human and animal health professionals as well as patients and the public; and (3) basic research of mechanisms of resistance and virulence, identification of new drugs' targets of action, development of more rapid, reliable diagnostic tests and more vaccines, and an increase in research appropriations at the federal level.

International programs such as SENTRY were founded on the principles of the ASM task force and complement federal/national programs and industry-sponsored studies. SENTRY remains a unique program providing important information at local, regional, national, and worldwide levels. It also provides early indications about the value of investigational drugs, some directed at serious enterococcal infections.

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