Recovery of Resistant Enterococci during Vancomycin Prophylaxis

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We report a case of a patient undergoing hemodialysis who developed a wound infection and subsequently bacteremia with a strain of vancomycin-resistant enterococcus identified as *Enterococcus gallinarum*. He had been receiving vancomycin prophylaxis before developing these infections. Both isolates were susceptible to ampicillin, rifampin, teicoplanin, and daptomycin (LY146032).

There are approximately 80,000 people receiving hemodialysis in the United States. Over the past several years, it has become common practice to prophylactically administer vancomycin to these patients. In many respects, vancomycin is ideally suited to this purpose, since skin flora, composed primarily of gram-positive bacteria, commonly infect these patients and vancomycin resistance in these organisms has only rarely been reported (9, 15). In addition, the drug may be given weekly during hemodialysis. Emerging resistance, however, is always a potential problem with the administration of prophylactic antibiotics. We report the first case of bacteremia with a vancomycin-resistant enterococcus recovered in the United States in a hemodialysis patient receiving prophylactic vancomycin. Susceptibilities to several newly developed antibiotics are also reported.

Case report. A 67-year-old man was admitted to the North Carolina Memorial Hospital on 6 September 1986 for evaluation of fever and swelling over his left femoral hemodialysis graft. Seventeen months before admission, the patient began hemodialysis. At that time, vancomycin (1 g intravenously [i.v.] each week) was administered. Vancomycin levels in serum between 14 and 21 μg/ml were reported on several occasions.

During the next year, his course was marked by repeated episodes of graft failure due to thrombus formation. On 12 June 1986, a left femoral graft was placed. One month later, swelling was noted around the graft site and an ultrasound of the area revealed several cystic collections.

Cultures of spontaneous drainage from one of these collections grew *Escherichia coli*, *Klebsiella pneumoniae*, and an *Enterococcus* sp. Broth macrodilution testing of the enterococcal isolate revealed that the vancomycin MIC was 16 μg/ml. The patient was maintained on his weekly vancomycin, and on 6 September he returned to the North Carolina Memorial Hospital after 48 h of fever and chills. His oral temperature was 100.2°F (ca. 37.9°C), and his physical exam was remarkable for a foul-smelling exudate from a wound over his graft site.

Enterococci were recovered from three of four blood cultures taken upon admission. Microdilution MIC testing (8) revealed that for this isolate, penicillin and ampicillin MICs were 1.0 μg/ml and the vancomycin MIC was 16.0 μg/ml. The vancomycin MIC was confirmed by broth macrodilution MIC testing (8). Kill-curve studies (11) with measured levels in serum of vancomycin (20 μg/ml) and gentamicin (5 μg/ml) demonstrated synergy for the blood isolate

(data not shown). Broth macrodilution MICs of other agents with antistreptococcal activity are reported in Tables 1 and 2.

The bacteremia of the patient was treated with 2 g of piperacillin i.v. every 8 h and 150 mg of gentamicin i.v. after each dialysis. His temperature rapidly returned to normal, and he was sent home to complete a 4-week course of vancomycin and gentamicin. The blood and wound-drainage isolates were sent to the Centers for Disease Control in Atlanta, Ga., for identification and confirmation of the vancomycin MIC findings. The blood and wound isolates were identified as *Enterococcus gallinarum*. The MIC of vancomycin for both isolates was 16 μg/ml.

The patient did well until 15 December 1986, when he again experienced rigors and fever. A golf-ball-sized mass was palpable in his left groin, and when it was aspirated, an *E. gallinarum* isolate for which the vancomycin MIC was 16 μg/ml was recovered. Four of four blood cultures, however, grew *Pseudomonas aeruginosa*, and his graft was removed. He did well after a 4-week course of ceftazidime and tobramycin.

Discussion. Vancomycin resistance in gram-positive organisms is highly unusual. It had been reported in only two genera of gram-positive organisms, *Leuconostoc* (12) and *Lactobacillus* (2, 17). The enterococci, which include the species *E. faecalis*, *E. durans*, *E. faecium*, and *E. avium*, have been reported to be susceptible to vancomycin, with MICs in the range of 0.5 to 8.0 μg/ml (1, 3, 5, 7, 10, 14). Recently Uttley et al. (16) have isolated eight vancomycin-resistant *Enterococcus* strains from the blood of patients with end-stage renal disease. There have been no previous reports of *E. gallinarum* of human origin. For the blood and wound isolates of enterococci reported here, the vancomycin MICs were 16 μg/ml.

The *E. gallinarum* isolates could be clearly differentiated from both *Leuconostoc* sp. and *Lactobacillus* sp. The MICs of vancomycin for both isolates were relatively low compared with those reported for *Leuconostoc* sp. (MIC range, 500 to >2,000 μg/ml). In addition, both *Enterococcus* isolates were able to split arginine and acidify litmus milk, characteristics which are not found in *Leuconostoc* sp. (6). Also, the *E. gallinarum* isolates failed to produce gas in *Lactobacillus* MRS broth, and *Leuconostoc* sp. produces gas in this broth (6). For certain lactobacilli, vancomycin MICs may be similar to those for the isolates described here. However, the Gram stain, colonial morphology, and definitive physiologic characteristics unique to *E. gallinarum* clearly indicate that these isolates were not lactobacilli.

It was clear that the blood and wound isolates were the

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TABLE 1. Broth macrodilution MICs and MBCs for
two enterococcal isolates

Drug	MIC/MBC[a] (μg/ml) for:	
	Blood isolate	Wound isolate
Vancomycin	16/>128	16/>128
Daptomycin	2/8	1/4
Teicoplanin	1/32	1/32
Rifampin	≤0.12/≤0.12	<0.12/0.5
Ciprofloxacin	4/8	4/8
Fusidic acid	2/>128	4/>128
Bacitracin[b]	32/128	16/64

[a] MBC was defined as 99.9% killing of the starting inoculum, which was between 1.2×10^5 and 1.4×10^5 CFU/ml.

[b] Values for bacitracin are given in units per milliliter.

same organism. Both were clearly enterococci: they were typed in serogroup D antiserum, were catalase negative and alpha-hemolytic, hydrolyzed bile-esculin, and grew in the presence of 6.5% NaCl and at 10 and 45°C. Biochemically, the isolates differed from other *Enterococcus* species by their motility and nonpigmentation. Only two species of enterococcus are motile, *E. gallinarum* and *E. casseliflavus*. *E. casseliflavus* is pigmented (yellow), but *E. gallinarum* is not (4). The *E. gallinarum* isolate had two typical physiologic characteristics of the type strain for this species (4).

The antimicrobial susceptibilities of the two isolates were similar (Table 1). For both isolates described here, ampicillin MICs were as expected for enterococci, but surprisingly, both were resistant to vancomycin. We do not know whether other *E. gallinarum* isolates have the same antimicrobial susceptibilities as these isolates. The most active agents by weight were rifampin, daptomycin (LY146032; an investigational lipopeptide made by Eli Lilly & Co., Indianapolis, Ind.), and teicoplanin. We have previously reported (13) that teicoplanin had activity superior to that of daptomycin against *E. faecalis*. For the two *E. gallinarum* isolates described here, the MICs of those two agents were similar. Daptomycin, however, had bactericidal activity superior to that of teicoplanin (Tables 1 and 2). Variations in inoculum size did not significantly affect the activities of vancomycin, daptomycin, or teicoplanin (Table 2).

The patient was treated unsuccessfully with vancomycin and gentamicin, despite in vitro testing which predicted synergy of these two agents; ultimately, graft removal was

TABLE 2. Effect of inoculum size on antimicrobial activity
of vancomycin, teicoplanin, and daptomycin
against two enterococcal isolates

Drug	Inoculum (CFU/ml)	MIC/MBC[a] (μg/ml) for:	
		Blood isolate	Wound isolate
Vancomycin	10^2	16/16	16/>128
	10^4	16/>128	16/>128
	10^6	16/>128	16/>128
Teicoplanin	10^2	1/8	0.5/16
	10^4	1/16	0.5/128
	10^6	1/32	2/>128
Daptomycin	10^2	2/8	2/2
	10^4	2/8	2/8
	10^6	4/8	2/8

[a] MBCs for the 10^2-CFU/ml inoculum were defined as the lowest drug concentration allowing no growth on the subculture.

required. It is likely that the infection of this graft with a vancomycin-resistant enterococcus contributed to the failure of the graft. In this case, long-term vancomycin prophylaxis appears to have provided the selective pressure to produce an organism which was susceptible to penicillin but resistant to vancomycin. This lends credence to the contention that vancomycin prophylaxis leads to colonization and infection with vancomycin-resistant organisms. An additional case report has documented a vancomycin-resistant *Staphylococcus haemolyticus* strain which was isolated from the peritoneal dialysis fluid of a patient with end-stage renal disease and peritonitis (13). That patient had received several courses of vancomycin, both as therapy and as prophylaxis. MICs for his isolates gradually increased over several months of vancomycin therapy. Although widespread vancomycin resistance among gram-positive bacteria has not been reported, populations on long-term vancomycin-suppressive therapy, like those receiving hemodialysis, may prove to be a reservoir for these difficult-to-treat infections.

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