

Identification of a Tn1546-Like (Type 2) Element in Vancomycin-Resistant *Enterococcus faecium* Isolated from Hospitalized Patients in Japan

Vancomycin-resistant enterococci (VRE) have been isolated from about 50 Japanese patients since the first incidence in 1996 (1, 6; N. Fujita, M. Yoshimura, T. Komori, K. Tanimoto, and Y. Ike, Letter, Antimicrob. Agents Chemother. 42:2150, 1998). A connection between Japanese outbreaks and imported chickens has been suggested (5; Y. Ozawa, T. Nomura, T. Murata, K. Tanimoto, S. Fujimoto, and Y. Ike, Abstr. 1st Intl. Am. Soc. Microbiol. Conf. Enterococci, abstr. 103, p. 62, 2000). The link between animal colonization with VRE and human infection has been established in Europe, where the glycopeptide avoparcin previously was used as a feed additive (13). Clones of VRE identical to VRE of animal origin have been found not only in hospitalized patients but also in non-hospitalized humans, indicating the dynamic transfer of vancomycin resistance affecting public health (8, 12). Vancomycin resistance disseminates not only clonally but also by transfer of resistant elements horizontally to different clones of enterococci in animals and humans (4). Therefore, molecular epidemiological studies of these resistant elements are very important for the understanding of possible dissemination and elucidation of the spread of VRE.

In May 1998, a patient colonized with VanA-type *Enterococcus faecium* (VREF) was hospitalized at the Okayama University Hospital (Okayama, Japan). Fourteen isolates with indistinguishable pulsed-field gel electrophoresis (PFGE) patterns were collected from this patient, who was undergoing a bone marrow transplantation. The genetic background for vancomycin resistance was confirmed to be *vanA* (Tn1546) by using PCR methods (3, 9). A more thorough investigation of the Tn1546-like elements in these isolates was performed by using a combination of PCR, nucleotide sequencing, and hybridization (7). The results were compared to previously published variations in Tn1546 and indicated that all isolates possessed the unique molecular variations characteristic of a Tn1546-like (type 2) element, as defined by Jensen et al. (7; L. B. Jensen, Letter, Antimicrob. Agents Chemother. 42:2463–2464, 1998). The variations included the presence of an IS1216V-IS3-like element in the left end of Tn1546 and a point

mutation (G to T) at position 8234 in the essential *vanX* gene of Tn1546. We have further characterized Tn1546-like elements from selected VanA-type VRE of human and poultry origin from Japan. VRE have not been isolated from domestic porcine origin and imported pork samples tested in Japan (Y. Ike [Gunma University School of Medicine, Japan], personal communication). No Tn1546-like (type 2) elements were detected in *vanA*-containing VRE (7 *E. faecalis* and 1 *E. faecium* isolates) isolated from chicken fecal samples from three poultry farms (H. Yoshimura, M. Ishimaru, Y. S. Endoh, M. Suginaka, and S. Yamatani, Letter, Antimicrob. Agents Chemother. 42:3333, 1998). The Tn1546-like (type 2) element was detected in only 3 isolates (all from one hospital) out of 17 *vanA*-containing VRE (16 *E. faecium* and 1 *E. faecalis* isolates) isolated from 17 patients in six hospitals. The Tn1546-like (type 1) element was detected in 12 isolates from human and poultry origin, and 10 isolates indicating new variations in the Tn1546-like elements were untypeable according to the typing method of Jensen et al. (Table 1). Four human isolates possessing Tn1546-like (type 2) elements had closely related PFGE patterns, indicating clonal relationship according to the criteria of Tenover et al. (11), but were unrelated to other VRE isolates of human and poultry origin from Japan.

The Tn1546-like (type 2) element has previously been associated with VREF isolated from pig feces in Europe (2, 10, 14, 15) and has thus far been found in some hospitalized patients but not in feces from other food animals (Table 1). Genetic characterization of VRE isolates from patients in Japan revealed that Tn1546-like (type 2) elements were present in independent isolates from a small geographical area in Japan. Since Tn1546-like (type 2) elements have not been found in VRE of poultry origin either from Europe or from Japan, it is unlikely that the VRE containing Tn1546-like (type 2) elements originated from poultry. In Japan, no glycopeptides have been used as growth promoters for domestic pigs. Therefore, the Japanese VREF isolates containing Tn1546-like (type 2) elements could have come from porcine origin outside Japan.

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TABLE 1. VRE (*vanA* type) of human and animal origins from Japan, Europe, and the United States

Source	No. of isolates examined	No. of isolates originating from:			No. of isolates that were ^a :							
		Japan	Europe	United States	Subtype 1	Subtype 2	Subtype 3	Subtype 4	Subtype 5	Subtype 6	Subtype 7	Untypeable
Humans	50	18 ^b	27	5	11	7	6	8	2	2	1	13
Pigs	8		8			8						
Poultry	21	8 ^c	13		17						1	3
Total	79	26	48	5	28	15	6	8	2	2	2	16

^a Subtypes were determined according to methods described previously (7).

^b Out of 18 human isolates from Japan, 6 type 1 isolates, 4 type 2 isolates, and 8 untypeable isolates were found. Fourteen isolates with indistinguishable PFGE patterns from Okayama University Hospital were counted as one isolate.

^c Out of 8 poultry isolates from Japan, 6 type 1 isolates and 2 untypeable isolates were found.

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