

Occurrence of Vancomycin-Resistant Enterococci in Pork and Poultry Products from a Cattle-Rearing Area of France

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Meat products were collected from public retail outlets and tested for the presence of vancomycin-resistant enterococci (VRE) in an area with a high prevalence of VRE reported in human fecal samples. VRE were detected in 66% of the samples, and a predominance of VanC strains was found, which is also true for human fecal samples.

In a previous study we reported the high prevalence of vancomycin-resistant enterococci (VRE) in fecal samples from hospitalized (37%) and healthy nonhospitalized (11.8%) subjects living in the same local community (6). We found a predominance of *Enterococcus gallinarum* VanC strains (70.7%), together with 10.8% *E. casseliflavus* VanC strains and 18.5% *E. faecium* VanA strains, without any detectable VanB strains. A partial explanation for such high VRE prevalence was the use of a sensitive test with an enrichment broth step. Nevertheless, it was also suspected that the high levels might be related to the fact that subjects and patients were recruited from a predominantly agricultural area where avoparcin had been used as a growth promoter in animal husbandry. Assuming therefore that the food chain might be a likely source of VRE contamination in humans (1–3, 8, 9, 12), we investigated the occurrence of VRE in meat collected from local retail outlets.

During a 2-month period, between May and June 1999, 59 meat samples (50 pork and 9 poultry samples) were collected from butchers' shops, supermarkets, and other retail and wholesale outlets by environmental and veterinary health research laboratory staff. Approximately 25 g of each piece of meat was placed in 250 ml of buffered peptone water (bio-Mérieux, La Balme les Grottes, France) and homogenized with a "stomacher" (Waring blender; Poly Labo, Strasbourg, France). After incubation at 37°C for 24 h, 0.1 ml of the diluted sample was inoculated onto bile-esculin agar plates with and without 6 mg of vancomycin per liter and into bile-esculin broth supplemented with 4 mg of vancomycin per liter. Plates and broths were incubated at 37°C for 24 h. All esculin-positive broth cultures were subcultured onto bile-esculin agar plates with and without 6 mg of vancomycin per liter. Species identification of enterococci, susceptibility testing, and determination of glycopeptide resistance genotypes were performed as previously described (6). *E. faecium* strains with a *vanA* genotype were typed by pulsed-field gel electrophoresis (PFGE) as

previously described (6) and compared with *E. faecium vanA* strains of human origin isolated in our previous study (6).

We found high VRE contamination of the locally produced meat products, with 39 of the 59 samples (66%) being contaminated with VRE (9 poultry samples [9 of 9] and 30 pork samples [30 of 50; 60%]). This parallels the high prevalence of human VRE fecal colonization recorded in our previous study (6). Other European studies have observed VRE contamination in meat, from 8.3% of meat samples in Germany (10) to 79% of poultry samples in The Netherlands (14). Comparison of different data is hindered because the samples studied and detection methods vary considerably. However, all studies show the existence of a considerable pool of VRE in foods of animal origin (4, 10, 11, 14, 15). To our knowledge, only two studies are similar to our own, having effected a survey of human VRE fecal colonization and of VRE contamination in meat available at the retail level in one discrete geographical area (11, 14). One of these studies (carried out in The Netherlands) unexpectedly found a higher prevalence of VRE in poultry products (79%) (14) than in the fecal carriage of both hospitalized patients and patients living in the community (only 2%) (5). The other study (carried out in France) found a high VRE colonization rate both in humans (17%) (7) and in meat samples (41%) (11), though the levels recorded were lower than those found in our study.

Forty-nine VRE strains were isolated, 5 *E. faecium* strains (10.2%) with a *vanA* genotype, 1 *E. durans* strain with a *vanA* genotype, 29 *E. gallinarum* strains (59.2%) with a *vanC1* genotype, and 14 *E. casseliflavus* strains (28.6%) with a *vanC2* genotype. No *E. faecalis* strains and no strains with a *vanB* or *vanD* genotype were detected. All but two *E. faecium* strains were only isolated after broth enrichment. All *vanA* genotype strains were isolated from pork samples (Table 1). Poultry samples were all positive for VRE but only contained *vanC1* and *vanC2* genotype strains.

As in the human study (6), there was a predominance of VanC isolates and no *E. faecalis* strains were detected. This species distribution differs from that reported in other studies in which *E. faecium* and *E. faecalis* strains normally outnumber other species (4, 10–14). The proportion of *E. gallinarum* strains that we isolated is higher than that reported even by

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TABLE 1. VRE strain and genotype distribution in different meat products

Sample type	No. of samples with genotype:				% VRE (no. of VRE isolates/total)
	<i>E. gallinarum</i> <i>vanC1</i>	<i>E. casseliflavus</i> <i>vanC2</i>	<i>E. faecium</i> <i>vanA</i>	<i>E. durans</i> <i>vanA</i>	
Cooked pork products	4	4	1	0	53.8 (7/13)
Uncooked pork products	11	4	4	0	66.6 (16/24)
Pork meat	4	2	0	1	53.8 (7/13)
Poultry meat	10	4	0	0	100 (9/9)
Total	29	14	5	1	

other studies using enrichment methods (10, 12, 14). In both the French (7) and Dutch (14) studies, *E. faecium* occurred as the principal species in human as well as meat samples.

The PFGE patterns of *E. faecium vanA* isolates from animal foodstuffs are presented in Fig. 1. Nonhuman isolates generated heterogeneous PFGE patterns that were different from those obtained with human isolates in our previous study (6). It was not possible in our study to demonstrate a link between *vanA E. faecium* strains of human and animal origins.

The high VRE contamination level described in this study confirms our predictions since avoparcin had been employed by meat producers in the local area. As suggested in a previous study (6), the high VRE contamination observed in meat samples might explain the high prevalence of VRE colonization in humans, particularly since enterococcus species distribution is comparable in both studies. However, no link was observed for *E. faecium vanA* strains. Further analysis of *E. gallinarum* and

E. casseliflavus strains by PFGE and analysis of transposons encoding high-level glycopeptide resistance of *vanA* strains might be able to shed light on possible links between human and animal VRE strains.

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1 2 3 4 5 6 7 8 9 10 11 12 13 14

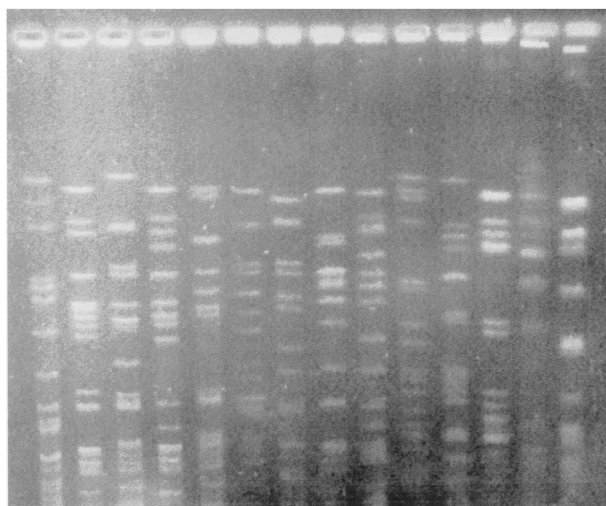


FIG. 1. *Sma*I restriction endonuclease patterns obtained by PFGE for *E. faecium vanA* strains from the following samples. Lane 1, cooked pork product; lanes 2, 3, 4, and 5, uncooked pork products; lane 6, patient 5 (control group); lane 7, patient 6 (control group); lane 8, patient 1 (control group); lane 9, patient 3 (hematology unit); lane 10, patient 8 (hematology unit); lane 11, patient 7 (hematology unit); lane 12, patient 4 (hematology unit); lane 13, patient 9 (hematology unit); lane 14, patient 2 (hematology unit).