

High-Frequency Recovery of Quinupristin-Dalfopristin-Resistant *Enterococcus faecium* Isolates from the Poultry Production Environment

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The occurrence of resistance to the streptogramin quinupristin-dalfopristin in *Enterococcus faecium* isolates from chickens on the Eastern Seaboard, was evaluated. Quinupristin-dalfopristin resistance was found in 51 to 78% of *E. faecium* isolates from the food production environment. The high level of resistance in this organism suggests that this reservoir of resistance may compromise the therapeutic potential of quinupristin-dalfopristin.

Vancomycin has largely been reserved to treat multiresistant *Enterococcus* sp. infections and methicillin-resistant *Staphylococcus aureus* (MRSA) infections; however, resistance has become so prevalent over the past 10 years as to present a crisis situation. Faced with a potentially untreatable pathogen, the U.S. Food and Drug Administration (FDA) has approved use of the streptogramin quinupristin-dalfopristin (Synercid) to treat infections caused by vancomycin resistant enterococci (VRE). Reports from German health agencies suggest that human carriage of quinupristin-dalfopristin-resistant *Enterococcus faecium* occurs at a rate of 14% (11). Although resistance to quinupristin-dalfopristin has been observed in the United States, there is insufficient information, as yet, to determine the incidence in the nonhospitalized community. These observations illustrate that a pool of resistance to this antibiotic can be found in the human microbiota. In the absence of selective pressure through clinical administration of the drug, the means by which these isolates acquired resistance has yet to be fully clarified. One possible source of resistance development may have been the use of an analogue of quinupristin-dalfopristin, virginiamycin, in poultry production. Virginiamycin has been used in the United States for more than 25 years to control clostridial diseases and subtherapeutically to promote the growth of commercial poultry. As part of a larger study examining multiresistant enterococci from poultry in a region of the Eastern Seaboard, we have discovered an unusually high prevalence of quinupristin-dalfopristin resistance in *E. faecium* isolated from chickens.

Sixty-seven samples were collected from poultry transport containers (PTC), litter samples, and cloacal swabs. Swabs taken from nine PTC from six farms were used to inoculate colistin-nalidixic acid (CNA) agar plates. Seventeen poultry

floor litter samples from 14 different farms were added at a 1:4 dilution to nalidixic acid-brain heart infusion-salt (NABS) enrichment broth for incubation at 35°C and were subsequently streaked onto CNA agar plates (5). Forty-one cloacal swabs were rinsed in tryptic soy broth (TSB) and spread plated onto either CNA or cephalixin aztreonam arabinose (CAA) agar (3) supplemented with 10 µg of quinupristin-dalfopristin/ml and 4 µg of ampicillin/ml. Colonies suspected to be *Enterococcus* spp. were isolated and identified using a modification of the protocol described by Facklam and Collins (2). Susceptibilities of *E. faecium* isolates to quinupristin-dalfopristin were assayed using the Kirby-Bauer disk diffusion methodology on Mueller-Hinton agar (with resistance defined as a zone of >15 mm) or by use of the Sensititre antimicrobial testing system with a breakpoint MIC of >2 µg/ml (7, 8).

Twenty-one (51.2%) of 41 *E. faecium* isolates from cloacal swabs taken from chickens were resistant to quinupristin-dalfopristin. Of the 27 *E. faecium* isolates from PTC swabs and litter samples, 21 (77.7%) were resistant (Table 1). Variance in the observed quinupristin-dalfopristin resistance incidence is likely due to the differences in sampling methodology as well as to differences in colonization or carriage rates among flocks.

The high rate of recovery of quinupristin-dalfopristin-resistant *E. faecium* from chickens in the United States was troubling, but not surprising, given the routine application of the selective agent virginiamycin to poultry feed. Welton et al. have reported that virginiamycin-resistant *E. faecium* prevalence in domestic turkeys from Michigan can be as high as 100% because of the lengthy exposure of flocks to virginiamycin (10). Resistance development in chickens is significant in that the growout period for broiler chickens is 6 to 8 weeks versus the 16 to 19 weeks required for turkeys. In addition, per capita consumption of chicken products in the United States exceeds that of turkey by a factor of 4.3 (9).

Various plasmid-borne genetic elements, which carry quinupristin-dalfopristin resistance in agriculturally derived *E. faecium*, have been demonstrated to cross into *E. faecium* isolates

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TABLE 1. Profile of susceptibilities of *E. faecium* isolates of poultry origin to quinupristin-dalfopristin

Sample type	No. susceptible/total no. (%)	No. resistant total no. (%)
PTC and litter	6/27 (22.2) ^a	21/27 (77.7) ^a
Cloacal swabs	20/41 (48.7) ^b	21/41 (51.2) ^b

^a Determined using broth microdilution (resistance, MIC of >2 µg/ml).

^b Determined using agar disk diffusion (resistance, zone diameter of >15 mm).

of human origin in vitro (4). This finding was unsettling because of the problems that hospital physicians currently encounter in controlling nosocomial VRE. While VRE with resistance to quinupristin-dalfopristin have not yet emerged as a problem in the clinical setting, it is possible that this resistance pattern will eventually occur. Indeed, the incidence of quinupristin-dalfopristin-resistant VRE has been reported to be 7.4% in German hospitals in 1998 (6). In fact, a recent report has shown that genes for quinupristin-dalfopristin resistance and vancomycin resistance were found on the same plasmid in an *Enterococcus* isolate from France (1).

Although the use of antibiotics at subtherapeutic concentrations in agriculture in the United States has not been definitively established as a contributor to the rising problem of antibiotic resistance in human clinical medicine, the presence in the food production environment of a population of *E. faecium* that harbors mobile resistance elements to quinupristin-dalfopristin is cause for concern. The European Union has banned the use of subtherapeutic antibiotics in agriculture as a precautionary measure due to a presumed risk to human health. The possibility that these strains may infect humans or that resistance elements may cross to the human microbiota, notably *E. faecium*, represents sufficient risk to warrant broader and continuous surveillance in the United States.

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