

VANCOMYCIN-RESISTANT ENTEROCOCCI (VRE) AS INTESTINAL FLORA FROM POULTRY IN RIYADH

S.M. Hussain Qadri, PhD, Dip. ABMM, FAAM; A. Gerald Postle, RT (CSLT); M. Qari, MD;
M.A.H. Quraishi, PhD, Dip. ABMM

Vancomycin-resistant enterococci (VRE) were first reported in France in 1988.¹ Since then, these organisms have been reported to have caused human infections in the USA, UK, Germany, The Netherlands, Spain and Saudi Arabia.²⁻⁵ The number of cases infected with VRE, as reported by the US Centers for Disease Control and Prevention, had jumped from 99 in 1992 to 278 in 1994, with a rapid increase in colonization, and a fecal carriage rate of 86%.⁶ Since colonized patients as well as domestic animals and environment can serve as significant reservoirs for human acquisition of VRE, we determined the presence of these organisms in sewage and fecal samples of poultry, camels, goats and sheep in Riyadh and its vicinity. Our findings are reported in this paper.

Materials and Methods

Treated sewage samples from Riyadh and Ha'ir and fecal specimens from animal (camel, sheep, goat) farms and poultry farms and outlets were collected in sterile plastic bottles and inoculated on 5% sheep blood agar (BAP), MacConkey agar and Campy BAP (containing 10 mg vancomycin/L). All plates were incubated at 35 °C for 24-48 hours. Suspected colonies of enterococci were selected from either BAP and/or Campy BAP, and identified by morphology, cultural characteristics and biochemical tests as per standard procedure and confirmed by the GPI (gram-positive identification) system (Vitek Systems, Inc., Hazelwood, MO, USA). Minimum inhibitory concentration (MIC) of *Enterococcus* against antimicrobials was determined by Vitek Systems and confirmed by standard broth dilution method.⁸ For MIC determination, Mueller-Hinton broth, pH 7.2-7.4, was used

as the test medium and inoculated with the turbidity-adjusted suspension of the isolate to achieve a final inoculum of 10⁵ CFU/mL.⁸ *E. faecalis* ATCC 29212 was used as a quality-control organism during MIC determination. Results were found to be within expected ranges.

Results

Two samples of treated sewage from Riyadh sewage treatment plant, four from the Ha'ir region, 32 fecal samples from animal farms and 26 from poultry farms/outlets were tested for the presence of VRE. None of the sewage samples or fecal material from camels, sheep, or goats grew VRE. Twelve (46%) of the 26 samples from poultry grew VRE. Four of these were identified as *E. faecium*, and eight as *E. gallinarum*. All 12 VRE had an MIC (mg/L) of >256 for vancomycin, >16 for ampicillin, >32 for cefazolin, cefuroxime and gentamicin, >64 for ceftriaxone, >2 for clindamycin and >8 for ciprofloxacin and erythromycin. All the isolates were susceptible to nitrofurantoin with an MIC of 4-16 mg/L. Only one VRE strain was resistant to trimethoprim-sulfamethoxazole with an MIC of 8/152 mg/L. All the isolates were also resistant to the glycopeptide avoparcin (MIC>256 mg/L), which is commonly used as an additive in animal/poultry feed.

Discussion

During the last two years a number of studies have reported the presence of VRE in the environment, animal husbandry, poultry and dog food.⁹⁻¹¹ Although only limited information is available regarding the possible connection between human VRE infections and potential environmental or animal reservoirs, Bates et al.⁹ found the same ribo-type among VRE isolates from human patients, sewage and pigs, suggesting the existence of an environmental or animal reservoir of VRE for human infections.

It is known that the selection for VRE takes place as a consequence of the therapeutic use of vancomycin in

From the Departments of Pathology, King Faisal Specialist Hospital and Research Centre (Drs. Qadri, Postle and Qari), and Jacobi Medical Center (Dr. Quraishi), Bronx, New York, USA.

Address reprint requests and correspondence to Dr. Qadri: King Faisal Specialist Hospital and Research Centre, Department of Pathology and Laboratory Medicine, MBC 10, P.O. Box 3354, Riyadh 11211, Saudi Arabia.

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hospitals and the use of antimicrobials as additives in animal feed. The US Food and Drug Administration allows the use of bacitracin, spiramycin, virginiamycin, flavomycin and the glycopeptide avoparcin as supplements in the animal and poultry feed. Recently, Aarestrup¹⁰ reported that 77% of the enterococcal isolates from poultry farms using the glycopeptide avoparcin (Avotan, Cyanamid International, Wayne, NJ) were resistant to vancomycin, whereas none of the isolates was resistant to vancomycin from ecological farms that did not use any antimicrobial supplement in the feed. Our attempt to find the ingredients in animal and poultry feeds met with total refusal from the management of these farms. We did find that large amounts of avoparcin is imported into the Kingdom, but the local distributor was reluctant to divulge the information about the buyers/recipients of this glycopeptide. However, in view of the earlier reports,^{9,11} it seems feasible that the occurrence of VRE in 12 of the 26 poultry samples may have resulted from the use of avoparcin in poultry feed. Although no breakpoints have been established for avoparcin, all 12 VRE isolates from poultry had an MIC >256 mg/L for avoparcin. Both *E. faecium* and *E. gallinarum*, which were isolated from poultry, are known causative agents of human disease.

So far human VRE infections are not a major problem in Saudi Arabia. To our knowledge, only six cases of VRE infections and some colonization have been seen or reported.^{4,5,7,10} As the prudent use of antimicrobials by clinicians is effective in lowering the emergence of resistant isolates in humans, animal husbandries and poultry farms should be discouraged to use avoparcin and other antimicrobial agents as feed supplement, in order to minimize acquisition of resistant flora.

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