Author's response to reviews

Title: Prevalence of Vancomycin-Resistant Enterococci colonization and its risk factors in chronic hemodialysis patients in Shiraz, Iran

Authors:
Mehrdad Askarian (askariam@sums.ac.ir)
Soheila Shaghaghian (shaghaghians@sums.ac.ir)
Ojan Assadian (ojan.assadian@meduniwien.ac.at)

Version: 2 Date: 9 May 2007

Author's response to reviews: see over
ANSWER TO REVIEWER COMMENTS

April 21 2007

MS: 2080882066127033

Referee 1

C1: Of 146 cultures (presumably 146 individuals, which the authors should clarify), 9 dialysis patients were at least transiently colonized in the rectum with VRE.
We have obtained 146 samples from 146 patients. Nine samples of 9 different patients were positive for VRE. We have clarified this issue now in the result section and the abstract of the manuscript.

C2: Risk factors included antimicrobial receipt within 2 months before culture and hospitalization during the previous year, although these factors only accounted for 2 and 3 patients, respectively; the authors should clarify whether this is a total of 5 patients or whether there is overlap in these numbers.
We thank the respected reviewer for pointing out this aspect. Indeed, there is an overlap in these numbers. Both patients having received antibiotics within the previous 2 months also have been hospitalized during the past year. The factor ‘Hospitalization during past year’ seems to be a surrogate for antibiotic consumption. We have added this information into the result section and have briefly discussed it in the discussion section of the manuscript.
While this aspect does not change the unimodale statistics, we do not calculate a P value for the combined factors, because the numbers we are dealing with are too small.

C3: The authors might clarify why they didn’t use broth enrichment to increase their yield and use vancomycin-containing selective and differential media from the outset, as many other groups have done for surveillance culture surveys.
The reason for not including an enrichment step was simple an organizational matter inherent to the investigated units. The respected reviewer is correct that using broth enrichment might have increased the yield in VRE. Studies have shown that vancomycin broth enrichment is superior to direct plating for the detection of VRE. Novicki et al. demonstrated in 2004 (J Clin Microbiol 2004; 42:1637-40.) that addition of a broth enrichment step leads to the detection of significantly more VRE isolates than direct plating alone. Yet, Reisner et al. (Infect Control Hosp Epidemiol 2000; 21:775-9) clearly showed that use of an enrichment broth medium is required to recover VRE contaminating environmental surfaces; however, direct inoculation to selective solid medium is adequate to recover VRE in patient perianal specimens.
The issue raised by the respected reviewer would have been of ultimate importance in the case if we would have reported that we did not find any VRE in the investigated population. The purpose of our study, however, was not to determine a precise estimation of the prevalence in hemodialysis patients, but to obtain enough information to calculate risk factors for VRE.
carriage. Furthermore, in an epidemiological sense pertaining to risk of transmission it remains debatable how relevant a VRE carrier is, if the isolate is only detectable by means of enrichment cultures.

We have discussed this issue briefly in the second paragraph of the discussion of the initial manuscript (lines 11-17). Based on the reviewer’s recommendation we have, however, extended the discussion on the consequences.

C4: There are no results of molecular typing (e.g., pulsed-field gel electrophoresis) of the VRE; such assessment of the relatedness of the strains would be needed provide a better understanding of the epidemiology of VRE in these dialysis units. Obtain molecular typing (e.g., PFGE) of the strains.

As requested, we have added molecular biological results. The respected reviewer suggested performing a PFGE; because we already have a very well established protocol for random-amplified polymorphic DNA (RAPD) PCR, we performed the initial typing by using our RAPD-PCR method as described by Barbier et al. earlier. The RAPD typing technique is quicker and easier to perform, but has disadvantages both in reproducibility and discriminatory power. However, because the typing result showed that all clones are distinguishable, we therefore believe that in this case performing the PFGE – clearly the gold standard – might not be necessary anymore. We sincerely hope that the respected reviewer finds this approach acceptable.

We have included a figure of the typing results together with a brief interpretation of the results.

C5: The abstract has more detail than is necessary for such a short paper and could be abbreviated considerably.

The abstract was shortened by approximately 20% (from 239 words to 198).

C6: It appears that all of Table 1 can be incorporated in Table 2. (Table 1 is solely the addition of the two columns in Table 2 and therefore the two can be combined).

We have followed the respected reviewer’s comment accordingly. The two tables are now combined to one table.