Author's response to reviews

Title: Improved primers for the detection and identification of human enterovirus 71 by RT PCR

Authors:

   David Perera (davidperera@yahoo.com)
   Yuwana Podin (ypodin@yahoo.com)
   Winnie Akin (winnie_akin@yahoo.com)
   Cheng-Siang Tan (kennytcs@hotmail.com)
   Dr Mary J Cardosa (janecardosa@yahoo.co.uk)

Version: 3 Date: 7 Apr 2004

The revisions we have made are as follows:

1. Additional references in the Background section as suggested by Dr. Rombaut.

2. Included identification of specimens yielding the PCR products shown in Figure 4, as suggested by Dr Shimizu. For consistency, we have also done the same for Figure 1. This additional information is included in the legends of the 2 figures.

3. A few typographical errors pointed out by the reviewers have been corrected.

4. The compulsory revision required by Dr Shimizu has not been done because the point of this work was really to document the problem of misidentification of CVA16 as HEV71 using the 159S/162A primers. This problem is a recent problem and we felt it important for virologists in the field to learn of the problem. In doing the sequencing to sort out why this mispriming was happening, it was only logical to use the new information to design primers for our own use and we now wish to share these with the community. Our intention is not to compare these new primers with those of other groups which are not generally used. There is no doubt that the 159S/162A primers have been instrumental in making it possible to rapidly identify HEV71 outbreaks and are used by numerous groups in the region and elsewhere. We have changed the title of the manuscript to make the objective of this paper more clear. We have also changed the final paragraphs of the discussion to reflect this point. We have removed all speculative comments about primers which we have not directly tested. However, we want to stress that this work is not meant to cast any doubt on the fact that the 159S/162A primers have been extremely valuable to all of us in the field.

5. We have removed the tables listing the virus isolates and the clinical specimens used and have put these into the supplementary materials as Additional files 1 and 2. These are pdf files. Table 4 in the original manuscript is now Table 2.

6. Due to the fact that the focus of this work really is CVA16 misidentification rather than HEV71 in isolation of CVA16, we believe that the data in Figure 6 is not only useful but essential to the case. We have thus not removed this figure.