Reviewer's report

Title: The Limitations of Real-Time PCR Amplification for the Detection of Invasive Salmonella serovars in Blood and Bone Marrow Specimens from Enteric fever Patients

Version: 2 Date: 23 September 2009

Reviewer: Nora Cardona-Castro

Reviewer's report:

Major Compulsory Revisions

My personal opinion is that the work continues having very big methodological limitations that the authors have to review in objectively way:

1. In consequence of the well information about S. Typhi and S. Paratyphi genes and their functions, product of important Salmonella research, is necessary to use this knowledge for the development of specific and sensitive PCR tests. The new concept used in science is: “from de bench to the field”. Based in this premise, the DNA sequences used as targets for the current real time PCR lack of this background. The use of specific genes that identify Salmonella serovars is a very good approach for development of PCR tests. The use of any DNA sequence found in a chromosome as target for a PCR test could be the reason of the low sensitivity of the test developed in current work.

2. Abstract does not include the sensitivity and specificity of the test.

3. The use of a simple PCR internal control warranties the availability of the test in any time and place.

4. I can not identify the positive control of the test used.

5. Calculation of the sensitivity and specificity of the test is still lacking of the right methodology.

6. There is a confusion of terms with respect to sensitivity. Sensitivity is used indistinctly for the minimal concentration of DNA detected by the test and for the percentage of true positive detected by the test.

7. The authors give a non acceptable reason to explain the fails of this work. They supported the fails of this work arguing that it was performed in a developing country. Unfortunately the science work in developing countries is more difficult, but in my own experience we can do acceptable and good quality research using standard procedures even though this reality.

8. Methods have to be directed in the right section no in results.
Level of interest: An article of importance in its field

Quality of written English: Acceptable

Statistical review: Yes, but I do not feel adequately qualified to assess the statistics.

Declaration of competing interests:

I declare that I have no competing interests