Reviewer's report

Title: Two stages PCR assay for detection of human brucellosis in endemic area

Version: 4 Date: 17 February 2013

Reviewer: Javier Guitian

Reviewer's report:

I appreciate the authors have considered and addressed comments made by the reviewers. However, I reiterate some of the points made in my previous review:

Major comment: the selection of individuals into the study has to be extremely clear for the readers. The authors write “all participants meeting inclusion criteria…” participants in what? What was the “source population”? all people seeking medical treatment in xxx hospitals from 1st January xxx, this has to be very clearly stated. Who assessed and how the inclusion criteria? When one of these individuals met the inclusion criteria, was this person automatically included in the study?

Minor essential comments:

The way your discussion is written suggests, in my opinion, that Saudi Arabia is a “developing country”, which I don’t think is the case.

You refer to the paper by Dean as “recording observations”, in fact this paper is a review, it does not present evidence from field observations, you should reword or find an alternative reference.

In your discussion you still fail to provide a robust explanation for the high proportion of people positive to both B. abortus and B. melitensis. You mention keeping livestock of different species as a reason, and it makes sense, but is this supported by data from the animal health services that shows mixed-species herds to be infected by the two Brucella species?

Level of interest: An article whose findings are important to those with closely related research interests

Quality of written English: Needs some language corrections before being published

Statistical review: No, the manuscript does not need to be seen by a statistician.