Reviewer's report

Title: Evidence of West Nile Virus Infection in Nepal

Version: 3  Date: 12 July 2014

Reviewer: Luisa Barzon

Reviewer's report:

This and interesting study which reports, for the first time, sequencing data on West Nile virus detected from two patients with fever from Nepal.

I recommend the below reported revisions to improve the quality of the results and to correct some data in the manuscript.

Major Compulsory Revisions

1. Abstract: line 40. Two sera (and not twelve) tested positive by RT-PCR (out of 14 with positive WNV antibodies by ELISA), as reported in the manuscript text.

2. Background: lines 50-60. Taxonomic classification of the Flavivirus genus and WNV lineages should be updated.

3. Background: lines 57-58. In the recent years, WNV lineage 2 has circulated also in Europe and Russia.

4. Background: lines 62-64. Kunjin virus is classified as WNV lineage 1b. Neutralizing antibodies cannot be distinguished by neutralization assays from those induced by other WNV strains.

5. Case Presentation: lines 121-122. How was secondary dengue infection diagnosed?

6. Case Presentation: lines 126. Please specify that the genomes had highest nucleotide sequence similarity with WNV lineage 1a.

7. Case Presentation: lines 126-127. For the shared genome sequence fragments, the percentage of nucleotide identity between the two genomes should be calculated.

8. Case Presentation: lines 128-131. For the long sequence contigs, especially those mapping in E and NS5, and for large sequences shared by the two genomes, phylogenetic trees should be generated including WNV lineage 1a sequences from GenBank. This analysis will provide important information on the phylogenetic relationship of the two WNV genomes from Nepal with other genomes reported worldwide.

9. Conclusions: lines 149-152. As above recommended, this finding should be supported by a phylogenetic tree analysis.
10. Conclusions: lines 149-152. How was the sequence similarity with WNV lineage 1 and WNV lineage 2 defined?

**Level of interest:** An article of importance in its field

**Quality of written English:** Not suitable for publication unless extensively edited

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

**Declaration of competing interests:**

I declare that I have no competing interests