Reviewer's report

Title: Mycobacterium tuberculosis ecology in Venezuela: Epidemiologic correlates of common spoligotypes and a large clonal cluster defined by MIRU-VNTR-24

Version: 2 Date: 4 May 2009

Reviewer: Viviana Ritacco

Reviewer's report:

The authors have answered my points sufficiently well to allow their manuscript to be published and I have no major compulsory revision to advise.

Minor Essential Revisions:

Below I copy paragraphs of the manuscript with suggested corrections/comments within triple square brackets.

1. Page 8

Epidemiologic data was not available for all of the patients from whom the 1298 strains had been isolated, but for several parameters the information was recorded for 25% of the 1298 patients. Overall, of those patients with available data, the mean age was 37.6 years, 71% were between 15 and 45 years of age, while 29% were over 45. Sixty nine percent were men, 74% had a [an acid fast bacilli smear??] positive clinical specimens, 88% had pulmonary disease, and 75% had cavitations according to the chest X-rays (data not shown). Venezuelans made up 93% of patients and foreign-borns just 7%.

2. Page 8

The distribution of spoligotype patterns in isolates from the foreign born population did not appear different from that of native Venezuelan patients. Comparisons of patients in different regions revealed that the average patient age was similar for the two urban areas – 39 years for Carabobo and 37.8 years for Caracas – and 36.8 years for the Amazon region (Table 2). However it was much younger, 32.6 years, for patients from the indigenous Delta Amacuro region. Also, while males made up about 71% of patients from Caracas, 70% from Carabobo [(69%) percentage duplicated??]]

3. Page 9

There were no significant differences when all patients in clusters were compared with all patients with non-clustered isolates. However, Table 3 shows that patients whose isolates had the most common spoligotype, SIT 17, were significantly younger [(mean age 34 years) repeated in next line]] compared to patients with other spoligotype strains (mean ages 34 vs 39 years, p = 0.0008). The SIT 17 patients also had lower mean ages when the comparisons were
stratified for each of the three regions with the most isolates, but the differences did not quite reach statistical significance (Caracas 35 vs 38.4, p = 0.077; Carabobo 34.5 vs 39.8 p = 0.076; and Amazonas 30.6 vs 39.1, p = 0.056).

4. Page 9
As seen in Table 4, patients with SIT 17 isolates were also more likely to have had clinical specimens positive for Acid Fast Bacilli (AFB) by microscopy when compared overall, or just within the Caracas cohort, but the differences were only significant compared to other Caracas patients (SIT 17 vs non-SIT 17, all patients: OR = 1.47, p = 0.14; Caracas: OR = 1.96, p = 0.04). Compared within the Caracas group, specimens from patients with the SIT 53 spoligotype were less likely to be AFB positive, but the difference was not statistically significant (OR = 0.68, p = 0.55).

5. Pages 27-29
Tables 2, 3 and 4 are illegible

6. Table 1 - Geographic Distribution of the Most Common Spoligotypes in Venezuela
Spoligotypes found in at least isolates, ranked by number of isolates, with SIT,...........

7. Figure 3 – Dendrogram of Venezuelan M. tuberculosis strains
Dendrogram combining MIRU-VNTR and Spoligotype results from representative strains generated by Bionumerics software (Applied Maths) using the unweighted pair group method of averages (UPGMA). MIRU patterns with 3 or fewer changes are considered clonal. Inside the box are the 605 strains defined as clonal by spoligo and by 24 MIRU- VNTR loci. Only MIRU 12 results were available for the strains of SIT 605 isolated in New York.

My advice is to accept the manuscript after minor essential revisions

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

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**Quality of written English:** Acceptable

**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