Author’s response to reviews

Title: Genotyping and clinical characteristics of multidrug and extensively drug-resistant tuberculosis from a tertiary care tuberculosis hospital in China

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Author’s response to reviews: see over
Dear Editor:

According to the comments and suggestions from the two reviewers and you, we have modified this manuscript (MS: 1204211201862874), entitled “Genotyping and clinical characteristics of multidrug and extensively drug-resistant tuberculosis from a tertiary care tuberculosis hospital in China”, and submit the revised manuscript for further consideration.

The point-by-point responses to the concerns about the manuscript (MS: 1204211201862874) as follow:

(1). Why did you select 15-locus panel?

**Authors’ response:** IS6110-restriction fragment length polymorphism (RFLP) has been used as the gold standard method for genotyping *Mycobacterium tuberculosis* since the early 1990s. However, this method is labor-intensive, requires weeks for culturing the isolates and subsequent DNA purification, and suffers from problems of interpretability and portability of the complex banding patterns. Recently, MIRU-VNTR, a simpler and faster genotyping method with discriminatory power equivalent to that of IS6110 RFLP, has been considered a good alternative to the standard method for studying the transmission dynamics of *M. tuberculosis*.

MIRU-VNTR genotyping is performed by amplifying a panel of 12, 15 or 24 loci. Compared to the 12-loci version, 15-locus (6 loci from the 12-loci version and 9 new ones) and 24-locus panels have higher discriminatory power and a better correlation with RFLP data. The 15-locus panel was considered to have the highest efficiency because it contained 96% of the resolution obtained with the whole 24-locus system in the study by Philip Supply et al. The 15-locus panel is proposed as the standard for routine epidemiological discrimination of *M. tuberculosis* isolates, and the 24-locus system as a high-resolution tool for phylogenetic studies. Obviously, the 15-locus panel is less labor-intensive than the 24-locus system.
(2). What do you mean by single patients?

**Authors’ response:** “A total of 804 *M. tuberculosis* clinical isolates were obtained from single patients with pulmonary TB living in Jiangxi province.” mean “A total of 804 *M. tuberculosis* clinical isolates were obtained from an equal number of different patients (each isolate from each patient) with pulmonary TB living in Jiangxi province”.

(3). Define what HGDI is?

**Authors’ response:** The Hunter–Gaston discriminatory index (HGDI) was proposed by Hunter and Gaston to evaluate the discriminatory ability of typing systems. HGDI can be derived from elementary probability theory and is given by the following equation:

$$HGDI = 1 - \frac{1}{N(N - 1)} \sum_{j=1}^{s} x_j(x_j - 1)$$

Where $N$ is the total number of isolates in the typing method, $s$ is the number of distinct patterns discriminated by MIRU-VNTR, and $n_j$ is the number of isolates belonging to the $j$th pattern.

This index indicates the probability of two strains sampled randomly from a population belonging to two different types. Thus, a HGDI value of 1.0 would indicate that a typing method was able to distinguish each member of a strain population from all other members of that population. Conversely, an index of 0.0 would indicate that all members of a strain population were of an identical type. An index of 0.50 would mean that if one strain was chosen at random from a strain population, then there would be a 50% probability that the next strain chosen at random would be indistinguishable from the first.

(4). Define previously treated patients? Is it default, relapse or treatment failures?
Authors’ response: According to WHO (World Health Organization), previously treated patients were defined as patients who have received 1 month or more of anti-TB drugs in the past, may have positive or negative bacteriology and may have disease at any anatomical site. It is important to identify previously treated patients because they are at increased risk of drug resistance. Previously treated patients are further classified as default, relapse or treatment failures group by the outcome of their most recent course of treatment (shown in Table 1 below). At the start of therapy, specimens should be obtained for culture and drug susceptibility testing from all previously treated patients. Treatment depends on whether the patient has relapsed or is returning after default or after prior treatment has failed.

Table 1 Subgroup among previously treated patients by the outcome of most recent treatment

<table>
<thead>
<tr>
<th>Patients</th>
<th>Bacteriology</th>
<th>Outcome of most recent prior treatment</th>
</tr>
</thead>
<tbody>
<tr>
<td>Previously treated</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Relapse</td>
<td>+</td>
<td>Cured</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Treatment completed</td>
</tr>
<tr>
<td>Failure</td>
<td>+</td>
<td>Treatment failed</td>
</tr>
<tr>
<td>Default</td>
<td>+</td>
<td>Defaulted</td>
</tr>
</tbody>
</table>

a “+” indicates positive smear, culture or other newer means of identifying *M. tuberculosis*.

b “Cure” indicates a patient whose sputum smear or culture was positive at the beginning of the treatment but who was smear- or culture-negative in the last month of treatment and on at least one previous occasion.

“Treatment completed” indicates a patient who completed treatment but who does not have a negative sputum smear or culture result in the last month of treatment and on at least one previous occasion.

“Treatment failed” indicates a patient whose sputum smear or culture is positive at 5 months or later during treatment. Also included in this definition are patients found to harbour a multidrug-resistant (MDR) strain at any point of time during the treatment, whether they are smear-negative or –positive.

“Defaulted” indicates a patient whose treatment was interrupted for 2 consecutive months or more.
Finally, we appreciate the two reviewers (Dhammika Magana-Arachchi and Adane Mihret) for their comments and giving us good suggestions!

Also, many thanks for your kindness in processing this manuscript!
Thank you very much!

Best wishes,

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