Open Forum Infectious Diseases

RESPONSE TO LETTER TO THE EDITOR

Reply to Adzemovic et al

To the Editor—We thank Adzemovic and colleagues for their correspondence and interest in our article. The points raised make for interesting discussion.

We agree that it would be illogical that "response to therapy" be part of any tuberculous meningitis (TBM) definition as patients may deteriorate or even die despite tuberculosis (TB) treatment and non-TB causes of meningitis may improve in the absence of treatment. The question brought to light a blunder in our Methods section (related to a very early nonapproved protocol version that slipped into the document during edits and version changes). This was due to a transcriptional glitch on the OneDrive multiuser interface. We have now corrected this in the current online version of the manuscript [1] and are grateful to Adzemovic and colleagues for unearthing the error. The University of Cape Town Research Ethics Committee–approved protocol was used to carry out this study and that methodology is currently reflected in the corrected manuscript version. We have, for verification purposes, provided a copy of that protocol to the Editor, and this is available upon request through the corresponding author.

We strongly agree that symptomatic persons presenting with a meningitic syndrome in a high-burden setting with trace-positive Xpert Ultra cerebrospinal fluid (CSF) results should always be treated, and this is our clinical practice when faced with this scenario. The study-specific prevalence of TBM can vary in the African setting and will depend on a number of factors including type of health system, referral pathways, level of expertise of the attending clinician, level of the healthcare setting, patient access to care including affordability and socioeconomic status, service model (payment vs nonpayment), etc. Indeed, several studies have found a study-specific disease prevalence of 1% to approximately 20% [2].

We do not agree with the contention that CSF samples that are trace positive on Xpert Ultra are always a true positive and that they are invariably indicative of a falsely negative Sanger sequencing result. We contend that a CSF Xpert Ultra trace result might be a true positive or a false positive. The latter, for example, might be due to primer-dimers or other irrelevant DNA sequences, or a false-positive fluorophore-specific signal detection error (like the high interreader variability with a faint result line when using a lateral flow assay). We are not suggesting that CSF trace is related to previous TB as it is sputum [3, 4]. It is well known that results are stochastic and the false positivity rate is higher around a dichotomized cutpoint dividing positive from negative, and this principle applies to any diagnostic test. Indeed, of the 20 trace-positive CSF samples, in 12 of 20 (60%) the status changed when repeat Xpert Ultra testing was performed (6 samples changed from trace to "not detected"; 6 changed from trace to "very low"; 8 remained trace). Other tests like T-SPOT-TB (latent TB infection test; Revvity) deal with this problem by designating an "uncertainty zone" around the cutpoint where results should be regarded as unreliable [5–7]. Other polymerase chain reaction (PCR)–based TB assays such as Roche Cobas MTB and BD Max MDR-TB, which appear to have a similar sensitivity as Xpert Ultra [8], do not have this issue as they do not have a "trace" readout, which is typically at high cycle threshold values at the very limit of detection. Indeed, specificity dropped by 2.7% overall with Xpert Ultra introduction when compared to the MTB-RIF assay [4], implying a higher level of false positivity related to the trace readout.

Of the 20 trace CSF results, 10 were indeterminate on sequencing—that is, DNA was insufficient in quality or quantity to generate readouts. We cannot be sure how many of these were true positives or false positives. Eight sequencing results confirmed a *Mycobacterium tuberculosis* sequence. However, 2 showed non–*M tuberculosis* sequences (false positives). To maximize the chances of adequate amounts of DNA for the sequencing process, we also reamplified the PCR product in about a third of samples. There are also other lines of evidence that suggest that some trace results are likely false positives. For example, in several studies only about 10%–20% of sputum trace results were culture positive [9] (it is widely accepted that culture is more sensitive than Xpert Ultra).

In summary, we are unsure what proportion of CSF trace results are true positives versus false positives; however, our data and several other lines of evidence suggest that at least some of these results are likely false positives. Recognition of this issue is useful for a few reasons including clinical management, devising solutions to this problem in future assay design, and developing newer diagnostic approaches that might include a combination of host and pathogen biomarkers as previously suggested by some of the coauthors of the letter [10].

Notes

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