Rapid molecular detection of tuberculosis

Zbinden, A C; Keller, P M; Bloemberg, G V
stem cells is the standard by which their pluripotent potential is evaluated.\textsuperscript{1-3} We believe that the intended application of human induced pluripotent stem-cell lines should determine the evaluation method. The issue of pluripotency becomes important for studies of the functional mechanism of reprogramming. However, a reproducible and rapid method to determine the quality of newly established human pluripotent stem cells is urgently required. Perhaps an epigenetic and gene-expression signature that selectively defines fully reprogrammed pluripotent stem cells can be identified, since the variation of teratoma results brings up the question of this assay’s value as a standard for proving pluripotency of human stem cells.\textsuperscript{4}

We tested our lines for the reactivation of endogenous pluripotency genes, the silencing of retroviral transgenes, and the capacity to form the three germ layers. In close collaboration with the pathology department, we are in the process of correlating the results of teratoma assays with the ability to generate functional cardiac myocytes. Although induced cardiomyocytes may replace pluripotent stem cells for disease modeling, this technique needs to be reproducibly established in human cells.

Alessandra Moretti, Ph.D.
Jason T. Lam, Ph.D.
Karl-Ludwig Laugwitz, M.D.
Deutsches Herzzentrum München
Munich, Germany
klaugwitz@med1.med.tum.de

Since publication of their article, the authors report no further potential conflict of interest.


Rapid Molecular Detection of Tuberculosis

\textbf{To the Editor:} Boehme et al. (Sept. 9 issue)\textsuperscript{1} report encouraging results on the use of an automated molecular test for Mycobacterium tuberculosis and resistance to rifampin (Xpert MTB/RIF). However, the population of patients with clinical tuberculosis who have negative cultures still poses a problem of interpretation, which was not discussed in the article. Among study patients whose samples were culture-negative but who had symptoms of tuberculosis, 29.3\% had positive results on the automated test; these patients made up 4.3\% of the total number of automated test–positive patients.

In such patients, tuberculosis that was detected by the automated test may have had a false negative culture because of low bacillary load or overgrowth, but the possibility of false positivity cannot be excluded. Furthermore, 23 patients with nontuberculous mycobacteria in culture were excluded from the analysis. In our site in Tanzania and in other African locations, nontuberculous mycobacteria are frequently found in culture,\textsuperscript{2,3} so the capability of the automated test to discriminate between tuberculosis and nontuberculoius mycobacteria, for which preliminary results have been encouraging,\textsuperscript{4} would be of great interest. More effort should be made in future studies to elaborate on these two groups, thus clearing an uncertainty regarding the performance of the automated test.

Norbert Heinrich, M.D.
Medical Center of the University of Munich
Munich, Germany
heinrich@lrz.uni-muenchen.de

Andrea Rachow, M.D.
Mbeya Medical Research Programme
Mbeya, Tanzania

Michael Hoelscher, M.D., Ph.D.
Medical Center of the University of Munich
Munich, Germany

Dr. Hoelscher reports receiving research grants from the European and Developing Countries Clinical Trials Partnership for the development of tuberculosis diagnostics in adults and children. No other potential conflict of interest relevant to this letter was reported.

TO THE EDITOR: Although resistance to rifampin and isoniazid usually occurs concomitantly, rifampin monoresistance is known and well documented in certain populations.1,2,3 When the automated test detects rifampin resistance, clinicians would have to consider further diagnostic and treatment options. First, would the strain be assumed to have multidrug resistance, given the high probability of concomitant resistance to isoniazid? In that event, treatment options would probably include the remaining first-line agents (ethambutol and pyrazinamide) plus a fluoroquinolone and an injectable antituberculosis agent.4 Clinicians may still prescribe isoniazid until resistance to the drug is excluded. Further testing with the use of either a traditional assay or the Genotype MTBDRplus assay would still be needed. Nonetheless, at least one or more second-line agents would be initiated early in the treatment of the strain of tuberculosis with at least partial resistance. However, patients with rifampin monoresistance may be unnecessarily exposed to the untoward effects of injectable agents for tuberculosis until the complete drug-susceptibility pattern is obtained. Additional detection of isoniazid resistance would substantially enhance the utility of this test.

Nitin Bhanot, M.D., M.P.H.
Allegheny General Hospital
Pittsburgh, PA
nitinbhanot@gmail.com

No potential conflict of interest relevant to this letter was reported.


TO THE EDITOR: Boehme et al. report that an automated test for tuberculosis had a sensitivity of 97.6% and a specificity of 98.1% for the correct identification of patients with rifampin-resistant tuberculosis. Even though the test greatly advances the direct detection of M. tuberculosis, correct identification of rifampin-sensitive strains is essential, since the false diagnosis of multidrug-resistant tuberculosis is deleterious for patients.1

We have identified a patient with rifampin-sensitive tuberculosis for whom automated testing of bronchoalveolar lavage falsely showed rifampin resistance. The M. tuberculosis bacterial load detected by the automated test was low. The curve pattern generated in the automated test resulted in the interpretation “rifampin resistance detected.” In contrast, rpoB sequencing,2,3 line-probe testing (AID Diagnostika), repeated culture, and phenotypic drug-susceptibility testing revealed the presence of fully susceptible M. tuberculosis. In this patient, short-course therapy resulted in a clinical response.

A major limitation of the design of the automated test is that interpretation relies solely on a decreased level (or the absence) of wild-type beacon hybridization, rather than additional hybridization to mutant probes, which would add to the specificity of the test. Our results indicate that positive results for rifampin resistance on the automated test must be viewed with caution and should be confirmed by phenotypic or additional genotypic methods when possible.

Andrea Zbinden, M.D.
Peter M. Keller, M.D.
Guido V. Bloemberg, Ph.D.
University of Zurich
Zurich, Switzerland
bloomberg@immm.uzh.ch

No potential conflict of interest relevant to this letter was reported.

TO THE EDITOR: The study by Boehme et al., which reports a high degree of accuracy for an automated test for the identification of tuberculosis in adults, could revolutionize the diagnosis of tuberculosis globally. However, in high-burden settings, up to 30% of tuberculosis cases occur in children. The diagnosis in this age group is challenging, especially in the high-risk groups for disease progression and poor outcome, which include children who are under the age of 5 years.