

Short Communication

The role of the Xpert MTB/RIF assay among adolescents suspected of pulmonary tuberculosis in Rio de Janeiro, Brazil

**Thania Luiza de Andrade Sieiro^[1], Rafaela Baroni Aurílio^[2], Elizabeth Cristina C. Soares^[3],
Silvia S. Chiang^[4] and Clemax Couto Sant'Anna^{[5],[6]}**

[1]. Centro de Saúde Vasco Barcelos, Secretaria Municipal de Saúde de Nova Iguaçu, Nova Iguaçu, RJ, Brasil.

[2]. Instituto de Puericultura e Pediatria Martagão Gesteira, Universidade Federal do Rio de Janeiro, Rio de Janeiro, RJ, Brasil.

[3]. Centro Administrativo São Sebastião, Secretaria Municipal de Saúde do Rio de Janeiro, Rio de Janeiro, RJ, Brasil.

[4]. Department of Pediatrics, Alpert Medical School of Brown University, Center for International Health Research, Rhode Island Hospital, Providence, USA.

[5]. Departamento de Pediatria, Faculdade de Medicina, Universidade Federal do Rio de Janeiro. Rio de Janeiro, RJ, Brasil.

[6]. Rede TB de Pesquisa, Rede Nacional de Pesquisa em Tuberculose, Rio de Janeiro, RJ, Brasil.

Abstract

Introduction: The *teste rápido molecular para tuberculose* (TRM-TB) was introduced in 2014 in Brazil for tuberculosis screening. However, its role in adolescents in Brazil has not been studied. **Methods:** A descriptive study of adolescents with suspected tuberculosis using National Laboratory software. **Results:** Of 852 (15.4%) suspected cases, 131 were positive by TRM-TB and 2% were resistant to rifampicin. Among TRM-TB-positive cases, 105 (91.4%) were culture-positive. Sixty-four of 96 samples were sensitive to rifampicin by TRM-TB; 11 were resistant to other drugs by drug sensitivity test (DST). **Conclusions:** Among suspected cases, 16% were diagnosed by TRM-TB, of which 17% were drug-resistant by DST.

Keywords: Tuberculosis. Adolescents. Polymerase chain reaction.

According to World Health Organization (WHO) data, of the 30 highest tuberculosis (TB)-burden countries accounting for 87% of cases worldwide, Brazil ranks 20th in the number of new TB cases annually¹. In 2015, 68,000 new cases were recorded in Brazil, an incidence rate of 33.6/100,000. In the City of Rio de Janeiro, the TB incidence rate was 91/100,000 inhabitants, one of the highest nationwide (ECC Soares: unpublished data).

Molecular diagnosis of TB was introduced in Brazil through the *Xpert MTB-Rif system* (Cepheid, Sunnyvale, U.S.A.). [Teste rápido molecular para tuberculose (TRM-TB)], which is based on real-time polymerase chain reaction (PCR). It is an automated, rapid, and easy laboratory test that simultaneously detects *Mycobacterium tuberculosis* (*Mtb*) and rifampicin resistance (RIF) in approximately two hours. In October 2009, the TRM-TB was authorized for use in Brazil, and the WHO recommended its use in TB-endemic countries in December 2010²⁻⁴. In Brazil, bacteriological confirmation increased with use of the TRM-TB; however, its incorporation in the public health network since 2014 has not yet affected the global notification of TB cases in Rio de Janeiro⁵.

While the performance of the TRM-TB has been evaluated in adults and children, to the best of our knowledge, no studies have focused on its use in adolescents. In general, children have paucibacillary TB, which allows bacteriological confirmation in only 20% of cases. After the age of 10, patients begin to have bacilliferous forms of TB similar to those found in adults, making bacteriological diagnosis more feasible³. Therefore, we hypothesized that the TRM-TB would detect a higher proportion of adolescent TB cases compared to that in younger children but a lower proportion compared to that in adults.

We performed a cross-sectional study of adolescents, defined as individuals between 10 and 18 years of age, meeting the following inclusion criteria: 1) suspected of having pulmonary TB (PTB), 2) receiving care in the Primary Health Care Clinics in the municipality of Rio de Janeiro, Brazil, between January 1 and December 31, 2015, and 3) had sputum, induced sputum, and/or gastric aspirate samples analyzed by TRM-TB. We reviewed the detection of *Mtb* and RIF resistance by the TRM-TB in these samples. Eligible adolescents were identified from a database created from the Gerenciador de Ambiente Laboratorial (GAL), national laboratory software for the monitoring and controlling of laboratory exams implemented by the Ministry of Health of Brazil (MoH)⁶.

We described the frequency of the detection of *Mtb* and RIF resistance by the TRM-TB. We also report the Mycobacteria Growth

Corresponding author. Prof. Clemax Couto Sant'Anna.

e-mail: clemax01@gmail.com

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Indicator Tube (MGIT) culture results for *Mtb* and conventional drug sensitivity test (DST) to anti-TB drugs. The data are presented as frequencies and percentages.

Ethical considerations

This study was approved by the Instituto de Puericultura e Pediatria Martagão Gesteira da Universidade Federal do Rio de Janeiro Research Board (Proceeding 961.452 of 02/24/2015) and by the Municipal Health Secretariat of Rio de Janeiro (Proceeding 1.629.126 de 07/08/2016).

In 2015, in the municipality of Rio de Janeiro, 852 TRM-TB exams of spontaneous sputum, induced sputum, or gastric aspirate in adolescents were registered in the GAL platform. Of these, 824 were spontaneous sputum, 22 were induced sputum, and six were gastric aspirates. *Mtb* was not detected in 716 (84%) of the 852 samples and the test was non-conclusive in two samples (0.23%). *Mtb* was detected in 134 (15.7%) samples by TRM-TB, of which 131 (98%) and 3 (2%) were RIF-sensitive and RIF-resistant, respectively. Of the three adolescents with samples resistant to RIF by TRM-TB, one had a negative culture [microbial growth <20 urine flow cytometry (UFC)] and the other two had positive cultures. One of the two was also resistant to isoniazid by conventional DST (MGIT).

Culture was performed on 105 samples from among 131 adolescents with RIF-sensitive *Mtb* detected by TRM-TB. Ninety-six of these cases were culture-positive (91.4%). Conventional DST was performed on 64 of the culture-positive samples. Eleven (17%) were resistant to other drugs: 6 (9%) to streptomycin, three (5%) to isoniazid, and two (3%) to RIF and isoniazid. The distributions of the TRM-TB results, *Mtb* culture, and DST are illustrated in **Figure 1**.

TRM-TB is indicated for adolescents in Brazil suspected of having PTB. The study of TB among adolescents is difficult because disaggregated data for this age group seldom exist. The literature suggests that childhood TB comprises individuals <15 years of age; older individuals are grouped with adults. The lack of adolescent-specific reporting has led to the invisibility of the needs of this age group.

Most adolescents (≥ 10 years) have bacilliferous PTB, unlike the paucibacillary PTB seen in children <10 years^{3,7}. A high sensitivity (around 95%) of the *Xpert* is only reached in patients with bacteriologically confirmed TB⁸. The detection of *Mtb* in 16% of suspected adolescent PTB cases by TRM-TB is similar to the findings of a study of adults in Rio de Janeiro⁹. Unsurprisingly, our *Mtb* detection rate was higher than the 10.5% *Xpert* rate reported by Raizada et al.¹⁰ in India among 4,600 children (0-14 years of age) suspected of having TB. In this paper, *Xpert* was performed under optimum collection and transportation conditions and its sensitivity was double that of smear microscopy. Similarly, in a hospital-based study, Nicol et al.¹¹ assessed children and adolescents <15 years, of whom 24% were HIV-infected. The *Xpert* positivity among the hospitalized TB suspects was 13%. In contrast, Bunyasi et al.¹² investigated children <4 years of age in South African hospitals, reporting an *Xpert* positivity rate of 1.2% among subjects with

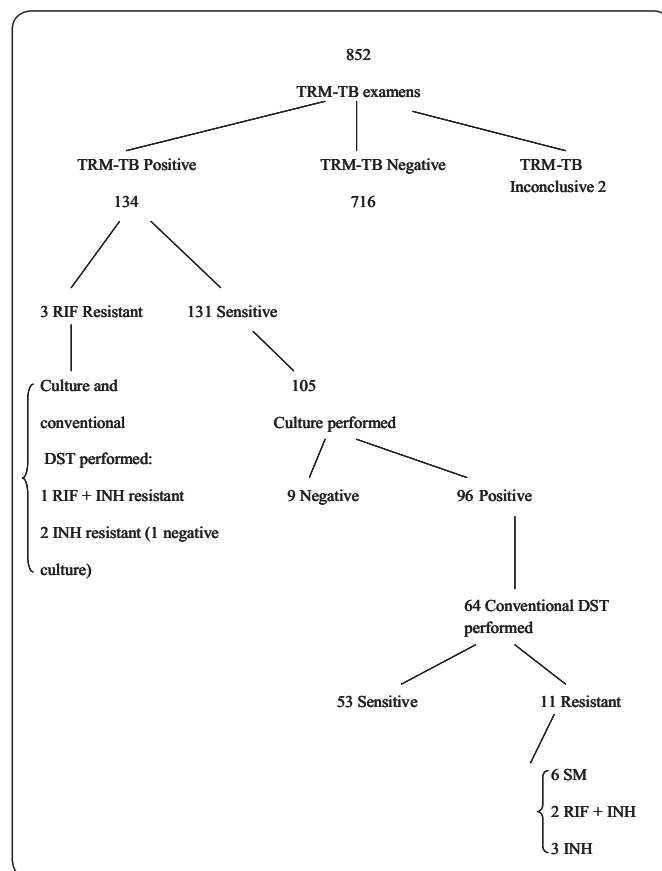


FIGURE 1: TRM-TB, culture, and drug sensitivity test results in 852 examinations of suspected adolescent tuberculosis. **TRM-TB:** *Teste rápido molecular para tuberculose*; **RIF:** rifampicin; **DST:** drug sensitivity test; **INH:** isoniazid; **SM:** streptomycin.

suspected TB. This finding is unsurprising because TB forms are predominantly paucibacillary in this age group. In another study of adult TB suspects, the *Xpert* positivity ranged from 13-30% and increased with TB prevalence in the study setting¹³.

In the present study, it was not possible to compare the TRM-TB and smear microscopy results. The guidelines from the MoH indicate that smear microscopy should only be requested for diagnosis in individuals who had been previously treated for TB¹⁴. However, a study in Rio de Janeiro did not show concordance between the smear microscopy and TRM-TB results in children⁵.

In our study, 98% of samples from adolescents with detectable TRM-TB results were sensitive to RIF, consistent with the low level of resistance of *Mtb* to RIF in Brazil. This finding was confirmed by the high detection of sensitivity to RIF by conventional DST. Of the 64 of 105 samples that were RIF-sensitive by TRM-TB, conventional DST showed resistance to other drugs in eleven (17%) samples. The most common was resistance to streptomycin, a drug widely used previously in Brazil. Among the 64 suspicious cases in which TRM-TB genotypically detected RIF-sensitivity and in which conventional DST was performed, two were phenotypically

resistant to RIF. Such individuals would go undetected by the TRM-TB. A possible reason for the discrepancy between conventional DST and *Xpert* is a mutation in sites not yet assessed by the TRM-TB^{13,15}.

Among the limitations of this study were the lack of clinical data and HIV test results of the adolescents with suspected TB. Therefore, we are unable to match the TRM-TB results with clinical diagnoses. In addition, we do not have conventional DST data for all samples in which *Mtb* was detected by the TRM-TB.

The TRM-TB may improve the diagnosis of PTB in adolescents. Since 25-30% of adolescents with PTB in Brazilian cities are negative by smear microscopy⁷, the use of TRM-TB may increase the number of TB-confirmed-cases. However, a negative TRM-TB result in adolescents does not eliminate the possibility of PTB. Clinical, radiological, and epidemiologic data are still very important in pediatric practice^{3,8}.

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Conflict of interest

The authors declare that there is no conflict of interest.

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REFERENCES

- World Health Organization (WHO). Country profiles for 30 high TB burden countries. Annex. WHO; 2017. 62p. Cited 2017 March 28. Available in: http://www.who.int/tb/publications/global_report/gtbr2017_annex2.pdf.
- Boehme C, Nicol MP, Nabeta P, Michael JS, Gotuzzo E, Tahirli R, et al. Feasibility, diagnostic accuracy, and effectiveness of decentralised use of the Xpert MTB/RIF test for diagnosis of tuberculosis and multidrug resistance: a multicentre implementation study. *Lancet*. 2011; 9776: 1495- 505.
- Ministério da Saúde (MS). Secretaria de Vigilância em Saúde. Manual de Recomendações para o Controle da Tuberculose no Brasil. Brasília: MS; 2011. 288p. Cited em 10 de março de 2017. Disponível em: http://bvsmms.saude.gov.br/bvs/publicacoes/manual_recomendacoes_controle_tuberculose_brasil.pdf
- Ministério da Saúde (MS). Secretaria de Ciência, Tecnologia e Insumos Estratégicos. Comissão Nacional de Incorporação de Tecnologias no SUS (CONITEC) - Relatório nº 49. Proposta de Incorporação do Xpert MTB/RIF como Teste para Diagnóstico de Tuberculose e para Indicação de Resistência à Rifampicina. Brasília: MS; 2013. 32p. Cited em 10 de março de 2017. Disponível em: <http://conitec.gov.br/images/Incorporados/TesteXpert--final.pdf>.
- Durovni B, Saraceni V, van den Hof S, Trajman A, Cordeiro-Santos M, Cavalcante S, et al. Impact of replacing smear microscopy with Xpert MTB/RIF for diagnosing tuberculosis in Brazil: a stepped-wedge cluster-randomized trial. *PLoS Med*. 2014;11(12):e1001766.
- Jesus R, Guimarães RP, Bergamo R, Santos LCF, Matta ASD, Paula Jr FJ. Sistema Gerenciador de Ambiente Laboratorial: relato de experiência de uma ferramenta transformadora para a gestão laboratorial e vigilância em saúde. *Epidemiol Serv Saúde*. 2013;22(3):525-9.
- Sant'Anna CC, Schmidt CM, March MFBP, Pereira SM, Barreto ML. Tuberculose em adolescentes em duas capitais brasileiras. *Cad Saúde Pública*. 2013;29(1):111-6.
- Chiang SS, Swanson DS, Starke JR. New diagnostics for childhood tuberculosis. *Infect Dis Clin North Am*. 2015;29(3):477-502.
- Soares ECC, Piller RB, Durovni B, Pio JE, Lourenço MC, Durovni P, et al. The use of the Xpert® MTB/RIF as a public health tool to treat MDR-TB cases timely – The first experience of its implementation in the City of Rio de Janeiro. *Am J Resp Crit Care Med*. 2016;193:A7693.
- Raizada N, Sachdeva KS, Nair AS, Kulsange S, Gupta RS, Thakur R, et al. Enhancing TB case detection: experience in offering upfront Xpert MTB/RIF testing to pediatric presumptive TB and DR TB cases or early rapid diagnosis of drug sensitive and drug resistant TB. *PLoS One*. 2014;9(8):e105346.
- Nicol MP, Workman L, Isaacs W, Munro J, Black F, Eley B, et al. Accuracy of the Xpert MTB/RIF test for the diagnosis of pulmonary tuberculosis in children admitted to hospital in Cape Town, South Africa: a descriptive study. *Lancet Infect Dis*. 2011;11(11):819-24.
- Bunyasi EW, Tameris M, Geldenhuys H, Schmidt BM, Luabeya AKK, Mulenga H, et al. Evaluation of Xpert® MTB/RIF assay in induced sputum and gastric lavage samples from young children with suspected tuberculosis from the MVA85A TB Vaccine Trial. *PLoS One*. 2015;10(11):e0141623.
- Luetkemeyer AF, Firnhaber C, Kendall MA, Wu X, Mazurek GH, Benator DA, et al. Evaluation of Xpert MTB/RIF versus AFB smear and culture to identify pulmonary tuberculosis in patients with suspected tuberculosis from low and higher prevalence settings. *Clin Infect Dis*. 2016;62(9):1081-8.
- Secretaria de Estado de Saúde. Nota técnica. Teste rápido molecular para tuberculose. Rio de Janeiro: Governo do Estado do Rio de Janeiro; 2014. 2p. Cited 2018 March 26. Disponível em: <http://www.riocomsaude.rj.gov.br/Publico/MostrarArquivo.aspx?C=0VeR8LQrFqE%3D>
- Pandey P, Pant ND, Rijal KR, Sherestha B, Kattel S, Banjara MR, et al. Diagnostic accuracy of GeneXpert MTB/RIF assay in comparison to conventional drug susceptibility testing method for the diagnosis of multidrug-resistant tuberculosis. *PLoS One*. 2017;12(1):e0169798.