ORIGINAL ARTICLE

String test: a potentially useful tool in the diagnosis of pulmonary tuberculosis in Brazilian children and adolescents

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ABSTRACT

This study investigated the potential use of the String Test (ST) for the diagnosis of pulmonary tuberculosis (PTB) in children and adolescents. This is a case series of patients aged 4-15 years presenting with clinically presumed PTB and submitted to ST in three pediatric TB referral centers in Brazil, between November 2017 and July 2020. The ST was performed in the morning, after 4-12 h of fasting, followed by ingestion of the capsule by the patient, which was attached to the patient’s malar region. The material was collected for simultaneous smear microscopy (acid-fast bacilli - AFB), culture and the molecular investigation by the GeneXpert MTB/RIF®. Thirty-three patients with presumed PTB were included and ST was performed in 26 (78.8%) of them and 7 (21.2%) patients could not swallow the cord. The diagnosis of PTB was established in 11 (42.3%) of the 26 patients who underwent the ST. The diagnosis of PTB was confirmed (by culture or GeneXpert MTB/RIF®) in 5 patients, 4 of whom were also positive by the ST. Two of them showed positivity by the GeneXpert MTB/RIF® only in the ST sample. Two other patients had a positive ST following the induced sputum test (AFB, GeneXpert MTB/RIF®, and positive culture in both specimens). Thus, ST was positive in 36.4% of the patients in whom PTB was diagnosed. ST could be a useful test for diagnosing PTB in children and adolescents.

KEYWORDS: Diagnostic techniques and procedures. Pulmonary tuberculosis. Children. String test.

INTRODUCTION

Pediatric tuberculosis (TB) accounts for 12% of all reported cases of TB globally and for approximately 1,200,000 new cases in 2019. Pediatric TB has been considered an underdiagnosed and underreported disease for many decades due to numerous factors, including the lack of specificity of symptoms, difficulty in obtaining samples for laboratory tests in children who frequently struggle to produce effective sputum, in addition to paucibacillary characteristics of pediatric TB1-3.

Given the difficulty in obtaining adequate clinical samples in children and adolescents, the string test (ST) might be a useful strategy. The ST was initially designed to evaluate enteric diseases and to isolate Giardia lamblia and Helicobacter pylori from gastric samples4. Over the last two decades, its efficacy in detecting Mycobacterium tuberculosis (Mtb) in gastric juice by retrieving the sputum swallowed by patients, similar to gastric lavage (GL), has been evaluated5. The ST in the pediatric population was well-tolerated by children, parents and the health care staff6.
The present study is a case series exploring the feasibility and performance of the ST, a noninvasive method, for obtaining clinical specimens for diagnosing pediatric pulmonary TB (PTB).

MATERIALS AND METHODS

This is a case series of patients with symptoms suggestive of PTB seeking medical care. They were prospectively included in the study by convenience sampling from three pediatric TB referral centers in three Brazilian State capitals (Rio de Janeiro, Curitiba and Sao Paulo), from November 2017 to July 2020. Children (aged <10 years) and adolescents aged 10-15 years treated as outpatients or inpatients were included. Patients who were unable to cooperate with the examination due to intellectual disability, difficulty swallowing or unstable clinical status were excluded.

The presumption of PTB was based on the presence of at least two of the following criteria: 1) symptoms compatible with TB: chronic cough, fever, weight loss and pneumonia that did not improve with antibiotic therapy; 2) chest radiograph with lymphadenopathy, cavitations, infiltrates, condensations, and/or miliary pattern, encompassing unilateral or bilateral changes; or 3) history of close contact with an adult diagnosed with PTB.

The ST was performed in the morning, in an outpatient clinic or ward after 4-12 h of fasting. Inpatients underwent other concomitant diagnostic tests for PTB in addition to the ST such as GL, induced sputum (IS) and bronchoalveolar lavage (BAL) according to the age of the patient and tests availability in the services. The decisions on what tests to perform and how many tests to perform were made at the discretion of the attending physician. The outpatient patients underwent the ST alone. Only one sample was obtained from each patient without repeating the ST on consecutive days. The clinical specimen collected for the ST was simultaneously submitted to at least one of the following diagnostic tests: acid-fast bacilli (AFB) smear, culture for Mtb and GeneXpert MTB/RIF®.

Patients with presumed PTB were allocated into the “confirmed TB” group if the culture or GeneXpert MTB/RIF® from any clinical specimen was positive.

The ST procedure in this study involved a capsule of approximately 1 cm in diameter attached to a thin, 80-cm long string. The material of the capsule is degradable in approximately 1 cm in diameter attached to a thin, 80-cm long string. The string was left inside the child’s stomach for 1 h and was then removed after rinsing the mouth with water. During this period, fasting was maintained and the participant was free to perform their usual tasks, including recreational activities. When the string was retrieved, the initial approximately 50 cm were cut and discarded. The remainder (around 30 cm) were stored in a Falcon tube with 2 mL of 0.9% saline solution to cover the string completely in the flask, and the sample was sent on the same day, within 2 h, to the mycobacteriology laboratory of the units participating in the study. The samples were then decontaminated with a 2% NaOH-NaCl solution. The material was processed for direct investigation of the presence of AFB, seeded in the culture medium, and submitted to the Xpert test, according to the manufacturer’s recommended procedure and the national guidelines issued by the Brazilian Ministry of Health®10. The teams responsible for performing the procedure were adequately trained through video classes, with communication about the services involved between their respective laboratories to clarify any doubts. Failure during the ST was defined as the inability to swallow the capsule after two attempts, according to the literature11.

A descriptive analysis of the data was performed using the Microsoft Excel® software (Microsoft, Redmond, WA, USA). All legal guardians of the patients signed an informed consent form before any procedure was performed. Patients aged 7-15 years have also provided a written assent to participate.

The Research Ethics Committee approved the present study in all the units participating in the study (Universidade Federal do Parana, approval Nº 61126816.1.0000.0096-2017 Curitiba, PR).

RESULTS

During the study period, 33 patients with presumed PTB were studied, as shown in Figure 1. Of the 33 patients selected, 21 (64%) were adolescents, and 2 (10%) of these 21 patients could not swallow the capsule. Of the 12 children (aged <10 years), five (42%) could not perform the ST, one of them due to a significant comorbidity (esophageal candidiasis) associated with AIDS.

Among the 26 patients with presumed PTB who were able to swallow the capsule for the ST, 18 (69%) were female, with a median age of 12 years (interquartile range [IQR]: 8.7-13.6). Seventeen (65%) of the 26 patients showed alterations in chest radiographs suggestive of PTB; the remaining 9 patients presented with symptoms and a
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33 patients admitted with clinical suspicion of PTB

26 (79%) were able to swallow the capsule for ST

7* (21%) could not swallow the capsule for ST

PTB confirmed
N=5 (19%)
(4* positive by ST and 3 positive by other techniques)

PTB not confirmed
N=6 (23%)
(all negative on ST and other techniques)

Withdrawn PTB
N=15 (58%)
(all negative on ST and other techniques)

Figure 1 - Flowchart of the inclusion of study participants. PTB = pulmonary tuberculosis; ST = String test; *5 children and 2 adolescents; **Of the 4 diagnosed by ST, in 2 of them the PTB diagnosis was established exclusively by the ST.

history of contact with a person with PTB. Of the 26 patients who underwent ST, 11 (42%), were diagnosed with PTB, with five confirmed as PTB and six with unconfirmed PTB. Of the 11 patients with TB who underwent ST, four (36%) were positive either by AFB, culture or GeneXpert MTB/RIF®. All were adolescents (aged 11-15 years), three with advanced radiological lesions suggestive of severe PTB, however one had a normal chest radiograph.

Table 1 summarizes the demographic, clinical, radiological and bacteriological characteristics of these 11 patients with PTB. In 15 patients who underwent ST, the diagnosis of PTB was ruled out. All of them showed

Table 1 - Demographic, clinical, radiological and bacteriological characteristics of the 11 studied participants with a diagnosis of pulmonary tuberculosis.

| Patient | Sex | Age (years) | Tuberculin skin test (mm) | Findings on the chest radiography | String test (AFB/culture/Xpert) | Other procedures (AFB/culture/Xpert) | HIV infection | Final diagnosis: PTB confirmed |
|---------|-----|-------------|---------------------------|---------------------------------|-------------------------------|-----------------------------------|---------------|---------------------------------|
| 01      | M   | 6.4         | 20                        | Adenopathy                      | Neg/Neg/Neg                   | Not performed                     | No            | No                              |
| 02      | F   | 12          | 10                        | Bilateral condensation and excavation | Neg/Neg/Neg                   | Sputum Pos/Pos/Pos               | No            | Yes                             |
| 03      | F   | 13.4        | 20                        | Adenopathy on the left          | Neg/Neg/Neg                   | Not performed                     | No            | No                              |
| 04      | F   | 11.8        | 20                        | Adenopathy on the right         | Neg/Neg/Neg                   | Not performed                     | No            | No                              |
| 05      | F   | 14.9        | 15                        | Condensation on the right       | Neg/Neg/Neg                   | Sputum induction Neg              | Yes           | No                              |
| 06      | F   | 12.1        | 20                        | Adenopathy on the right         | Neg/Neg/Neg                   | Not performed without information | No            | No                              |
| 07      | M   | 13          | 10                        | Bilateral condensation, infiltrate and bilateral excavation | Pos/Pos/Pos                   | Sputum induction Pos/Pos/Pos      | No            | Yes                             |
| 08      | F   | 13.3        | 10                        | Bilateral condensation, infiltrate and bilateral excavation | Neg/Pos/Neg                   | Sputum Pos/Pos/Not fulfilled      | No            | Yes                             |
| 09      | F   | 11.4        | 18                        | One-sided condensation         | Neg/Pos/Neg                   | Not performed                     | No            | Yes                             |
| 10      | M   | 9           | 0                         | Bilateral condensation         | Neg/Neg/Neg                   | Sputum and BAL Neg/Neg/Neg        | Yes           | No                              |
| 11      | F   | 15.7        | 21                        | Normal                         | Neg/Neg/Neg                   | BAL Neg/Neg/Neg                   | No            | Yes                             |

AFB = acid fast bacilli; BAL = bronchoalveolar lavage.
negative ST results, suggesting the procedure’s negative predictive value was high.

Among the seven patients who were unable to swallow the capsule, four (57%) were female, and the median age was 5.9 years (IQR: 5.4–9.9). One patient was diagnosed with unconfirmed PTB, and the others had the diagnosis of PTB ruled out. The patient diagnosed with unconfirmed PTB was a 13-year-old male adolescent who vomited after two attempts to swallow the capsule. His clinical and radiological parameters established the diagnosis of PTB. A 5-year-old female with AIDS could not swallow the capsule because of dysphagia secondary to fungal esophagitis, without vomiting or nausea. She was diagnosed with bacterial pneumonia on the basis of a clinical improvement with the use of antibiotic therapy for conventional pathogens. Other challenges reported were discomfort at the end of the first hour of the examination (two patients, a 6-year-old boy and an 11-year-old girl). There were no other complaints during the examination. At no time did the participants or their guardians ask to remove the string, nor was the string removed by the patient. No significant signs such as pain facies or other indications of discomfort or pain complaints during the test were reported by the patients, guardians, or observed by the health team in charge.

DISCUSSION

The present study was a Brazilian multicenter survey that sought to explore the feasibility and performance of using ST for obtaining clinical specimens from children and adolescents with presumed PTB. We evaluated recovery of Mtb from ST, comparing it with other diagnostic methods already established in the literature, including spontaneous sputum, gastric lavage (GL), induced sputum (IS), and bronchoalveolar lavage (BAL). Despite the small sample studied, the rate of success of this study (79%) was higher than that reported in the literature: 64% among adolescents, and 58% among children. In the study by Tafur et al., approximately 80% of the children aged 4-10 years could not swallow the capsule. The importance of this procedure is its applicability in children. Above this age, it is easier to obtain an adequate sputum sample, and their PTB is more similar to the adult pattern, making the Mtb isolation and PTB diagnosis easier.

ST studies in the HIV-infected pediatric population can have a great utility because of increased difficulties in diagnosing TB in such children. In HIV-infected patients, the clinical presentation of PTB can be highly heterogeneous. The tuberculin skin test (TST) is often nonreactive due to the patient’s immune system inability to react, and the radiological image is often atypical. Therefore, this population would undoubtedly benefit from such a test.

In the present study, no signs of intense discomfort or pain were observed and no participant removed the string before the pre-decided time, corroborating the findings of other studies that observed a good acceptance of the ST in the pediatric age group. Some authors have described that the moments of swallowing the capsule and removing the string were the least tolerable in children; however, the patients recovered quickly. In this study, four patients who underwent ST showed Mtb positivity in the smear microscopy, culture, or GeneXpert MTB/RIF. All were adolescents, three of them had advanced radiological forms of PTB, but the other patient had normal chest radiography results. Thus, the diagnostic contribution of ST on this occasion is still clear. The appropriate procedure duration in children to increase the sensitivity of ST is also still unclear in the literature. In some studies, the time with the string positioned intragastrically varied between 1 and 4 h. Considering that the patient should be fasting during the examination, 4 h would most likely reduce the compliance and efficacy of ST in children. Bae et al. used different durations for performing the ST in adults with sputum positivity for Mtb, and concluded that the time could be reduced to 60 min without compromising its effectiveness.

The present study has limitations including the small sample size and the collection of only one clinical specimen from each patient. This single sample might have contributed to the low positivity of ST in the samples studied. The literature shows that ST has a similar diagnostic accuracy of IS, GL, and culture. However, there are few studies that have compared ST with GL. The only report comparing both methods had a small sample size and showed similarity in the retrieval of Mtb in both techniques.

Some studies evaluated whether the ST can compete with other methods, especially IS and GL, for the retrieval of bacilli in children who cannot expectorate, in terms of accuracy, safety and acceptance. All methods require specific training of the healthcare staff, especially nurses. The disadvantage of ST is the inherent risk of bronchospasm in the patient, especially in children aged < 2 years, and the risk of transmission of bacilli via aerosols when the patient coughs. In our case series, there were no cases of bronchospasm and the acceptance was satisfactory. In general, the ST is an excellent tool to be applied in low-resource settings. The material to be examined can be prepared by the service staff as in this study and in previous studies from Peru. Although the costs for performing large-scale ST were not calculated in this study, it is assumed that it has financial advantages.
over other procedures like IS and GL. Additionally, the possibility of the ST being performed in an outpatient setting, as in the present study, reduces the hospitalization costs compared to GL. We thus point to the benefits of this technique as being outpatient-friendly, noninvasive and with no occupational hazards, compared to other available methods.

CONCLUSION

Increasing the sample size is necessary to obtain more robust data regarding the benefits of using this laboratory tool for diagnosing TB in the pediatric age group, thereby reducing costs related to hospitalization, morbidity and mortality in the affected population, even more if the specimen can be subjected to the Xpert Ultra, admittedly more sensitive than the GeneXpert MTB/RIF®.

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CONFLICT OF INTERESTS

None.

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