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Pulmonary Tuberculosis Diagnosed with Xpert® MTB/RIF Ultra in Stool Specimen in an Infant: A Case Report

İnfant Bir Hastada Dışkı Örneğinden Xpert® MTB/RIF Ultra ile Tanı Alan Akciğer Tüberkülozu: Vaka Sunumu

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ABSTRACT

Tuberculosis (TB) is an infectious disease spread by droplets containing Mycobacterium tuberculosis complex (MTBC) bacilli and is a significant cause of mortality and morbidity among the pediatric population. Although mycobacterial culture remains the gold standard for TB diagnosis, diagnosing TB in children is challenging due to difficulties in obtaining adequate respiratory samples, the typically low number of bacilli, and the long waiting time for culture results. Non-invasive diagnostic methods have advanced in recent years, including the Xpert® MTB/rifampicin (RIF) ultra assay, which is endorsed by the World Health Organization for diagnosing TB using samples such as stool and nasopharyngeal aspirates. We aimed to present a case report of a 10-month-old male patient diagnosed with pulmonary TB using the Xpert® MTB/RIF Ultra (Cepheid, United States of America) assay with stool samples. Despite the absence of acid-fast bacilli (AFB) in fasting gastric fluid, AFB was detected in stool samples, and MTBC was confirmed using the Xpert® MTB/RIF ultra system without rifampicin resistance. A culture of fasting gastric fluid eventually yielded MTBC in the MGIT960 culture system on the 18th day of incubation. Further analysis confirmed that the cause of the disease was M. tuberculosis. This case highlights the importance of combining rapid and sensitive diagnostic methods, such as Xpert® MTB/RIF ultra, for early detection and management of pediatric TB, especially in cases where sample collection is difficult.

Key Words: *Childhood tuberculosis; Stool; Xpert MTB/RIF ultra; Perinatal transmission; Demographic risk*

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ÖZ

İnfant Bir Hastada Dışkı Örneğinden Xpert® MTB/RIF Ultra ile Tanı Alan Akciğer Tüberkülozu: Vaka Sunumu

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Tüberküloz (TB), Mycobacterium tuberculosis complex (MTBC) basilini taşıyan damlacıklar ile yayılan ve pediatrik popülasyonda önemli bir mortalite ve morbidite sebebi olan infeksiyöz bir hastalıktır. Mikobakteri kültürü tüberküloz tanısında altın standart olmaya devam etse de çocuklarda tüberküloz tanısı koymak zordur. Bu zorluğun nedenleri arasında yeterli solunum yolu örneği almadaki zorluklar, genellikle düşük basil yükü ve kültür sonuçları için uzun süre beklenmesi yer alır. Son yıllarda özellikle çocukluk çağı TB tanısında invaziv olmayan örneklerde umut vadeden yeni gelişmeler bildirilmektedir. Dünya Sağlık Örgütü, çocuklarda TB tanısı için nazofaringeal aspirat ve dışkı örnekleri için Xpert® MTB/rifampisin (RIF) ultra (Cepheid, Amerika Birleşik Devletleri) testinin kullanımını onaylamıştır. Bu çalışmada Xpert® MTB/RIF ultra testiyle TB tanısı koyulan 10 aylık erkek hasta, perinatal bulaş şüphesi ve diğer demografik risk faktörlerinin güncellenmesi açısından sunulmaya değer görülmüştür. Hastadan alınan açlık mide sıvısında Asido-rezistan boyanan (ARB) basiller görülmemesine rağmen hastanın dışkı örneğinde ARB pozitif basiller tespit edilmiştir. Xpert® MTB/RIF ultra ile sadece dışkı örneğinde MTBC tespit edilmiş, RIF direnci saptanmamıştır. MGIT960 kültür sistemiyle, açlık mide sıvısı örneklerinden birinde inkübasyonun 18. gününde üreme tespit edilmiştir. Yapılan analizlerle hastalığın M. tuberculosis kaynaklı olduğu tespit edilmiştir. Sunulan olguda TB tanısı, dışkı örneği ile Xpert® MTB/RIF ultra sistemi kullanılarak hızlı ve kolay bir şekilde konmuştur. Bu sistemin, pediatrik vakaların hızlı tanısı ve tedavinin yönlendirilmesi açısından önemli bir gelişme sağlayacağı düşünülmektedir.

Anahtar Kelimeler: *Çocuk tüberkülozu; Dışkı; Xpert MTB/RIF ultra; Perinatal bulaş; Demografik risk*

INTRODUCTION

Tuberculosis (TB) is a contagious disease transmitted through the inhalation of infectious airborne particles generated by infected individuals. Children under five years of age are at greater risk of developing pulmonary TB[1]. The diagnosis of perinatal TB is often delayed as it can mimic other congenital diseases, including bacterial pneumonia[2]. TB diagnosis relies on culture, microscopy, and the GeneXpert *Mycobacterium tuberculosis* (MTB)/rifampicin (RIF) assay, which is recommended for the diagnosis of pulmonary TB in children by the World Health Organization (WHO). Gathering respiratory specimens from children can be challenging. However, considering that swallowed sputum contains mycobacterial DNA, the gastrointestinal tract renders stool a potential sample for TB confirmation using the GeneXpert MTB/RIF ultra assay[3]. Our aim in this article is to highlight the significance of stool samples as a potential option for the GeneXpert MTB/RIF ultra assay in the early detection of TB in children, particularly in cases where obtaining respiratory samples is challenging and there is a prolonged, undesirable waiting period for bacteriological confirmation through culture in pediatric patients.

CASE REPORT

A 10-month-old male patient was referred to our hospital with a disrupted onset of acute respiratory distress and cough. Physical examination revealed intercostal retraction and tachypnea. His laboratory findings, including blood urea, creatinine, sodium, potassium, calcium, serum albumin, alanine transaminase (ALT), aspartate transaminase (AST), and C-reactive protein (CRP) levels were within the normal range. Computed tomography revealed consolidations in the upper lobe of the right lung, as well as in the perihilar region and lower lobe of the left lung (Figure 1). It was noted that the patient had been hospitalized for two days with a diagnosis of bronchopneumonia one month prior to presentation. Furthermore, it was reported that the patient's mother had been diagnosed with tuberculosis and had commenced treatment seven

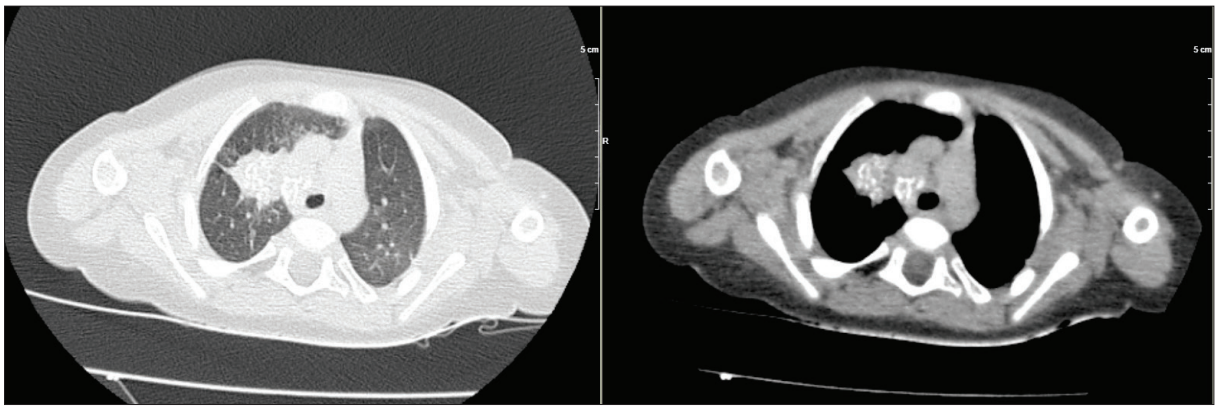

Figure 1. Lung computed tomography section of the case including the upper lobe of the right lobe.

months prior. The patient had received the Bacillus Calmette-Guérin (BCG) vaccine and exhibited a corresponding BCG scar. The tuberculin skin test result measured 8 x 6 mm. The tuberculin skin test results were negative for all members of the patient's family, including his mother, father, and five brothers. Anamnesis and imaging findings were evaluated together, and an investigation into the causes of pneumonia and *M. tuberculosis* was initiated immediately. Polymerase chain reaction (PCR)-based respiratory panel (Bosphore Respiratory Pathogens Panel Kit v8, Anatolia Geneworks) was performed using a nasal swab sample for rapid diagnosis. The assay results were negative for Coronavirus 229E, CoronavirusOC43, CoronavirusNL63, CoronavirusHKU1, parainfluenza virus 1, parainfluenza virus 2, parainfluenza virus 3, parainfluenza virus 4, human metapneumovirus (HMP), respiratory syncytial virus A/B, human bocavirus (HPoV), rhinovirus, enterovirus, adenovirus, human parechovirus (HPeV), pandemic influenza A – H1N1, *Legionella pneumophila*, *Bordetella pertussis* and *Haemophilus influenzae*. The assay revealed *Streptococcus pneumoniae* positivity. The patient was hospitalized. Initially, *S. pneumoniae* was considered a potential cause of pneumonia, and treatment with ampicillin-sulbactam treatment was initiated and continued for eight consecutive days. The subsequent interferon-gamma release assay (IGRA) result was positive. Obtaining respiratory specimens was particularly challenging for this patient. Therefore, on the day of hospitalization, gastric aspiration (over three consecutive days) and stool specimens were collected for culture and GeneXpert MTB/RIF assay to aid in diagnosis. Acid-fast bacilli were detected in the stool specimen, whereas gastric aspiration samples tested negative. Furthermore, the stool specimen was analyzed using the GeneXpert MTB/RIF system, yielding a positive result for the *M. tuberculosis* complex (MTBC). A fluorescent signal increase in the amplification curve can be observed in Figure 2. Consequently, the patient was diagnosed

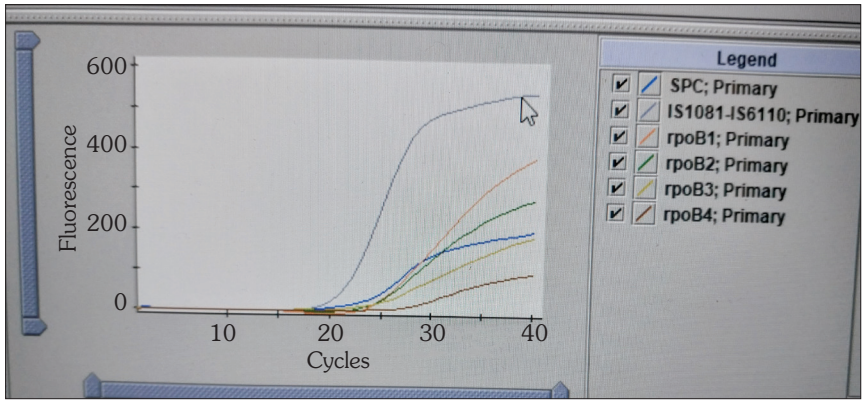

Figure 2. Fluorescence graph of analysis performed using GeneXpert MTB/RIF system from stool sample.

with pulmonary tuberculosis. To investigate the possibility of gastrointestinal tuberculosis, invasive procedures such as endoscopy or biopsy, were avoided due to the patient's young age and the associated risks. However, the clinical symptoms and radiological findings were consistent with pulmonary tuberculosis, providing sufficient evidence to establish the diagnosis. The genotypic analysis did not detect rifampicin resistance. The patient was admitted to the intensive care unit and placed in isolation as a precaution. Treatment was initiated with ethambutol, isoniazid, pyrazinamide, and rifampicin. On the 18th day of incubation, MTBC growth was detected in the gastric lavage sample inoculated into BACTEC MGIT 960 (Becton Dickinson, USA) liquid-based culture system. At the time of culture positivity, the patient had been receiving treatment for 15 days. Notably, there was a 15-day interval between PCR positivity from the stool sample and culture positivity of the fasting gastric lavage sample.

Differentiation between MTBC and nontuberculous mycobacteria (NTM) was performed using an immunochromatographic method (BD MGIT™ TBc identification test). To further distinguish MTBC species, the FluoroType® MTB assay (Hain Lifescience, Germany) was utilized, confirming the presence of *M. tuberculosis* in the patient. Phenotypic drug susceptibility tests were performed against streptomycin= 1.0 µl/ml, isoniazid (INH)= 0.1 µl/ml, RIF= 1.0 µl/ml, ethambutol (ETM)= 5.0 µl/ml and pyrazinamide (PZM)= 100 µl/ml using the BACTEC MGIT 320 system following the manufacturer's recommendations. On the eighth day of his hospitalization, the patient was referred to the hospital where he had previously been admitted for the continuation of his treatment.

DISCUSSION

TB is an infectious disease caused by *M. tuberculosis*, which can be life-threatening. If left untreated, the mortality rate can reach up to 50 percent[4].

TB remains the second leading cause of death from a single infectious agent, with more than 10 million people affected annually. Children aged 0-14 years account for 12 percent of all TB cases[5].

The clinical manifestations of TB include fever, weight loss, cough, and hemoptysis[6]. The radiological manifestations of TB vary and differ between studies. Computed tomography (CT) is useful to detect and interpret the lesions. Lymphadenopathy is the most common radiological feature of primary TB, while parenchymal consolidation is frequently observed. Cavitations may be observed in the advanced stages of the disease. Following treatment, parenchymal scarring may calcify, forming a Ghon focus. Upon reactivation of a tuberculosis lesion, patchy segmental consolidations can be observed in the apical and posterior segments of the upper lung lobes[7].

Dual infection with *S. pneumoniae* and *M. tuberculosis* is possible but relatively uncommon, particularly in the absence of significant risk factors such as immunosuppression. Reports indicate that *S. pneumoniae* infection can create an environment conducive to the reactivation of latent tuberculosis due to lung tissue damage and immune modulation. However, most documented cases of co-infection occur in TB-endemic regions or among immunocompromised individuals, such as those with HIV[8,9]. In our case, the patient lacked these predisposing conditions, making dual infection unlikely. Nevertheless, clinicians should maintain a high index of suspicion for coinfection in atypical presentations or regions where TB is endemic.

The risk of TB increases during pregnancy because of physiological immunological changes[10]. TB infection in pregnancy has negative outcomes, such as low birth weight, perinatal asphyxia, and even death[11]. In a study by Jonsson et al., 553 active TB cases were evaluated in 649.342 women who gave birth in Sweden between 2005 and 2013. Of these cases, 79 occurred postpartum (within 180 days of delivery), 85 occurred during pregnancy, and 389 occurred outside of pregnancy and the postpartum period. The overall incidence rates were 17% during the postpartum period, 12% during pregnancy, and 9% during the non-pregnant period. These findings demonstrate a significantly

increased risk of active TB during pregnancy and the postpartum period[12]. Separating the mother and baby is not always possible, especially in low-income settings. However, certain precautionary measures can be taken to reduce the risk of transmission, including ensuring adequate ventilation, wearing a mask while breastfeeding, sleeping in separate rooms, and maintaining these precautions for at least two weeks - until the mother is receiving adequate treatment or until no bacilli are detected on sputum microscopy[13]. The most common symptoms of TB infection in infants include fever, cough, and dyspnea, while the most frequently observed findings are rales and wheezing[14]. Mycobacterial culture is the "golden standard" for TB diagnosis[8]. The yield of mycobacterial culture for TB diagnosis in children is 30%-40%. However, culture-based diagnosis has certain limitations, as it is time-consuming, technically complex, and requires a tertiary-level laboratory. Therefore, patient history, clinical presentation, and radiographic findings are also crucial for TB diagnosis[15]. Apart from culture and smear microscopy, nucleic acid amplification-based assays, such as Xpert MTB/RIF or Xpert ultra, are recommended by the WHO for the diagnosis of tuberculosis, particularly in cases requiring immediate clinical decision-making and treatment initiation[16]. Xpert® MTB/RIF ultra (Cepheid, USA) is a cartridge-based nucleic acid amplification test designed for the rapid diagnosis of childhood tuberculosis and the detection of rifampicin resistance. Samples such as sputum, stool, and gastric aspirate can be tested using Xpert® MTB/RIF or Xpert® MTB/RIF ultra (Cepheid, USA)[17]. Perez-Risco et al. reported that the Xpert MTB/RIF ultra test had a specificity of 80% for identifying *M. tuberculosis* complex in stool samples[18]. A study by Kabir et al. showed 88.1% specificity and 58.6% sensitivity for Xpert MTB/RIF ultra in detecting MTBC in stool samples[3]. Both of the aforementioned studies align with the high predictability observed in our case, where the test supported the diagnosis of pulmonary tuberculosis.

The treatment regimen for a new patient with drug-susceptible pulmonary TB includes either a six-month regimen consisting of ETM, INH, PZA, and RIF (two months of ETM, INH, PZA, and RIF, followed by four months of INH and RIF) or a four-month regimen (two months of INH, PZA, RIF, and optionally ETM, followed by two months of INH and RIF) for patients aged between three months and 16 years[19]. It should be taken into consideration that the treatment of children under five years of age is more difficult[20].

As infants and young children are unable to produce respiratory secretions on demand, diagnosing TB can be challenging. However, the use of Xpert MTB/RIF systems with different clinical specimens overcomes the difficulties associated with the diagnosis of TB and early initiation of treatment. Our article demonstrates the significance of *M. tuberculosis*, one of the causes of pneumonia in children. Furthermore, the diagnosis made using GeneXpert MTB/RIF with a stool sample emphasizes the adequacy of this method where obtaining respiratory secretion is difficult.

While nucleic acid amplification tests, such as Xpert® MTB/RIF ultra, have revolutionized the rapid diagnosis of tuberculosis by enabling prompt initiation of treatment, culture remains the gold standard for definitive diagnosis. Therefore, despite the advances in molecular diagnostics, the role of culture remains central in the diagnosis and management of tuberculosis. Clinicians should be aware of the advantages and disadvantages of both methods and consider each patient as unique when determining the diagnostic approach.

CONFLICT of INTEREST

The authors declare that they have no conflicts of interest.

AUTHORSHIP CONTRIBUTIONS

Concept and Design: All of authors

Analysis/Interpretation: All of authors

Data Collection or Processing: All of authors

Writing: All of authors

Review and Correction: All of authors

Final Approval: CBK, GA

REFERENCES

1. *Churchyard G, Kim P, Shah NS, Rustomjee R, Gandhi N, Mathema B, et al. What we know about tuberculosis transmission: An overview. J Infect Dis 2017;216:629-35. https://doi.org/10.1093/infdis/jix362*
2. *Singh M, Kothur K, Dayal D, Kusuma S. Perinatal tuberculosis a case series. J Trop Pediatr 2007;53:135-8. https://doi.org/10.1093/tropej/fml074*
3. *Kabir S, Rahman SMM, Ahmed S, Islam MS, Banu RS, Shewade HD, et al. Xpert ultra assay on stool to diagnose pulmonary tuberculosis in children. Clin Infect Dis 2021;73:226-34. https://doi.org/10.1093/cid/ciaa583*
4. *Tiemersma EW, van der Werf MJ, Borgdorff MW, Williams BG, Nagelkerke NJD. Natural history of tuberculosis: Duration and fatality of untreated pulmonary tuberculosis in HIV negative patients: A systematic review. PLoS One 2011;6:e17601. https://doi.org/10.1371/journal.pone.0017601*
5. *World Health Organization (WHO). Global Tuberculosis Report, 2023. Available from: https://www.who.int/teams/global-tuberculosis-programme/tb-reports/global-tuberculosis-report-2023 (Accessed date: 19.08.2024).*
6. *Hopewell PC. A clinical view of tuberculosis. Radiol Clin North Am 1995;33:641-53. https://doi.org/10.1016/S0033-8389(22)00610-8*
7. *Andreu J, Cáceres J, Pallisa E, Martinez-Rodriguez M. Radiological manifestations of pulmonary tuberculosis. Eur J Radiol 2004;51:139-49. https://doi.org/10.1016/j.ejrad.2004.03.009*
8. *Wei M, Yongjie Zhao, Zhuoyu Qian, Biao Yang, Xi J, Wei J, et al. Pneumonia caused by Mycobacterium tuberculosis. Microbes Infect 2020;22:278-84. https://doi.org/10.1016/j.micinf.2020.05.020*
9. *Garcia R. Community-acquired pneumonia due to Streptococcus pneumoniae : When to consider coinfection with active pulmonary tuberculosis. Case Rep Infect Dis 2019;2019:1-4. https://doi.org/10.1155/2019/4618413*
10. *Tesfaye F, Sturegård E, Walles J, Bekele B, Bobosha K, Björkman P, et al. Dynamics of mycobacterium tuberculosis-specific and nonspecific immune responses in women with tuberculosis infection during pregnancy. Microbiol Spectr 2022;10:e0117822. https://doi.org/10.1128/spectrum.01178-22*
11. *Sobhy S, Babiker Z, Zamora J, Khan KS, Kunst H. Maternal and perinatal mortality and morbidity associated with tuberculosis during pregnancy and the postpartum period: A systematic review and meta-analysis. BJOG 2017;124:727-33. https://doi.org/10.1111/1471-0528.14408*
12. *Jonsson J, Kühlmann-Berenzon S, Berggren I, Bruchfeld J. Increased risk of active tuberculosis during pregnancy and postpartum: A register-based cohort study in Sweden. Eur Respir J 2020;55:1901886. https://doi.org/10.1183/13993003.01886-2019*
13. *Schaaf HS, Bekker A, Rabie H. Perinatal tuberculosis-an approach to an under-recognized diagnosis. Front Public Health 2023;11:1239734. https://doi.org/10.3389/fpubh.2023.1239734*
14. *Cruz AT, Starke JR. Clinical manifestations of tuberculosis in children. Paediatr Respir Rev 2007;8:107-17. https://doi.org/10.1016/j.prrv.2007.04.008*
15. *Basile FW, Nabeta P, Ruhwald M, Song R. Pediatric tuberculosis diagnostics: Present and future. J Pediatric Infect Dis Soc 2022;11:85-93. https://doi.org/10.1093/jpids/piac082*
16. *World Health Organization (WHO). Consolidated guidelines on tuberculosis. Module 3: Diagnosis - Rapid diagnostics for tuberculosis detection, 2021.*
17. *Kay AW, González Fernández L, Takwoingi Y, Eisenhut M, Detjen AK, Steingart KR, et al. Xpert MTB/RIF and Xpert MTB/RIF Ultra assays for active tuberculosis and rifampicin resistance in children. Cochrane Database Syst Rev 2020;8:CD013359. https://doi.org/10.1002/14651858.CD013359.pub2*
18. *Perez-Risco D, Rodriguez-Temporal D, Valledor-Sanchez I, Alcaide F. Evaluation of the Xpert MTB/RIF ultra assay for direct detection of mycobacterium tuberculosis complex in smear-negative extrapulmonary samples. J Clin Microbiol 2018;56:e00659. https://doi.org/10.1128/JCM.00659-18*
19. *World Health Organization (WHO). Consolidated guidelines on tuberculosis: Module 4: Treatment - Drug-susceptible tuberculosis treatment, 2022.*
20. *Fry S, Barnabas S, Cotton MF. Update on trends in childhood tuberculosis. Curr Opin Pediatr 2018;30:152-60.https://doi.org/10.1097/MOP.0000000000000581*

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