The authors present the design and implementation of associate investigations of young children with positive tuberculin skin test results. Case study analysis of an associate investigation was done using epidemiologic surveillance techniques, medical interviewing, sociogram mapping, tuberculin skin testing, radiographic evidence, and bacteriologic analysis. Deoxyribonucleic acid fingerprinting of the Mycobacterium tuberculosis isolates using a standardized IS6110-based restriction fragment length polymorphism analysis and IS6110-independent DNA spoligotyping methods was done to track and identify specific bacterial strains.

Deoxyribonucleic acid fingerprinting and spoligotyping done on isolates obtained from family members demonstrated same-strain transmission of M tuberculosis. Three adults with active pulmonary disease and six individuals with latent tuberculosis (TB) were discovered during this investigation. The arrival of a family member from Mexico who had the same strain suggests that the source case lives in Mexico.

A child with positive tuberculin skin test results indicates recent and potentially ongoing transmission of TB in the community. Targeted tuberculin skin testing performed on high-risk groups by primary care physicians allows for detection of TB infections. When TB infections are discovered in children, associate investigations can result in the discovery of undiagnosed adult cases and prevent further transmission within the community.

(Key words: tuberculosis, Mycobacterium tuberculosis, tuberculin skin testing, DNA, spoligotyping)

Tuberculosis (TB) continues to be a common problem faced by primary care physicians in the United States, despite the availability of effective preventive measures and treatment. The increased incidence of TB in the United States in the 1980s and early 1990s has been attributed to immigration from areas with a high prevalence of TB, human immunodeficiency virus–TB coinfection, and poor access to medical care. In the United States, the incidence of reported TB cases among foreign-born individuals is four to six times greater than that of the US-born population.

Primary care physicians play an important role in screening for latent TB infection. Tuberculin skin testing (TST) of foreign-born individuals and their children is an important measure for TB prevention. Primary care physicians should perform TST on all foreign-born patients who have immigrated in the past 5 years from countries in which TB is widespread. A child with TB indicates ongoing transmission of TB in the community, representing missed opportunities for prevention. Detection of TB infection in a child should lead to an associate investigation. Associate investigations, through the collaborative efforts of primary care physicians and public health departments, can lead to discovery of adults with TB.

The following case study illustrates the role of a primary care physician performing TST on a child, which led to the discovery of three adults with active pulmonary TB and six individuals with latent TB during an associate investigation.

Case study
MLM is a 1-year-old US-born daughter of Mexican parents residing in Tarrant County, Texas. She presented at a local health clinic for her 1-year well child examination, during which she received recommended immunizations as well as a TST. Evaluation revealed no overt signs or symptoms other than a nonproductive cough of 2 weeks’ duration. The TST developed a 16-mm induration 48 hours after placement. She was reported to the Tarrant County Health Department for further treatment.

Radiography revealed bilateral increased parahilar interstitial lung markings and left hilar adenopathy. The patient was admitted to the hospital to obtain early morning gastric aspirates for acid-fast bacilli (AFB) cultures. The gastric aspirate specimens were negative for AFB cultures and 8 weeks later were again negative. She was discharged from the hospital on directly observed therapy (DOT) regimen of isoniazid, rifampin, and pyrazinamide. An associate contact investigation
according to standard methods was initiated at the time of the report of positive TST results.\textsuperscript{5,9} Assessment of the physical and social environment revealed that MLM lived in a two-bedroom apartment with her parents and a maternal aunt. In addition, other extended-family members lived in the same apartment complex. Tuberculin skin testing was performed on all household contacts and family friends who had spent 8 hours or more per week with MLM. Six of the eight contacts found in the child’s immediate environment had positive TST results. In addition to skin testing, all contacts received chest radiographs and a comprehensive medical evaluation.

The child’s mother (patient A), who had lived in the United States for 2 years, reacted to tuberculin with an induration of 25 mm. She denied any symptoms of respiratory disease. Her chest radiograph showed evidence of cavitary infiltrates in her upper left lobe. On further questioning and after being told of the abnormality on her chest x-ray, she admitted to having had a “slight cold” for the past 4 months, which she considered insignificant. She related that these symptoms began after a 10-day visit with her father, who had been “taking medication for TB in Mexico for the past 4 years.” For this reason, she was believed to be at an increased risk of infection by drug-resistant organisms and was started on a regimen of isoniazid, rifampin, ethambutol hydrochloride, capreomycin, and pyrazinamide by DOT, pending results of sputum culture and drug susceptibility studies. Her initial sputum specimen analysis revealed greater than 10 AFB per high-power field. Cultures subsequently grew pansusceptible \textit{M tuberculosis}.

The child’s father reacted to tuberculin with an induration of 18 mm. He complained of a productive cough of 1 week’s duration, and his chest radiograph showed no evidence of pulmonary disease. He was also treated with a five-drug DOT regimen owing to the possibility of infection with a drug-resistant organism. Multiple sputum specimens were collected and were later negative for \textit{M tuberculosis}. He was then changed to treatment with isoniazid for latent TB infection.

The child’s 22-year-old aunt (patient B) had also lived in the United States for 2 years. She had positive TST results, and her radiograph showed cavitary infiltrates in her right upper lobe. She was also treated initially with a five-drug DOT regimen. Initial sputum specimens revealed the presence 1 to 10 AFB per high-power field. Cultures confirmed pansusceptible \textit{M tuberculosis}.

An uncle (patient C) arrived in the United States from Mexico 6 weeks after the start of the contact investigation. He also had positive TST results. Her radiograph showed cavitary infiltrates in her right upper lobe. She was also treated initially with a five-drug DOT regimen. Initial sputum specimens revealed the presence greater than 10 AFB per high-power field. His culture also grew pansusceptible \textit{M tuberculosis}. In his fourth month on a DOT regimen, he was arrested for public intoxication and deported to Mexico.

\textbf{Methods}

Starting in January 1996, as part of an ongoing Centers for Disease Control and Prevention study of the molecular epidemiology of TB, DNA fingerprinting has been performed on all \textit{M tuberculosis} isolates obtained from persons residing in Tarrant County with culture-confirmed TB. Patients A, B, and C were part of this study. Cultures for \textit{M tuberculosis} were performed by the mycobacteriology laboratory at Texas Department of Health using a Bactec radiometric system (Becton Dickinson, Paramus, NJ). Isolates were then analyzed by means of a standardized IS6110-based restriction fragment length polymorphism analysis method,\textsuperscript{10} and DNA fingerprinting of isolates obtained from each of the patients demonstrated identical five-band fingerprint patterns (Figure 1). As the discriminatory power of IS6110-based fingerprinting is diminished in strains with five or fewer bands,\textsuperscript{11} secondary fingerprinting of the isolates was performed by use of an IS6110-independent DNA fingerprinting method known as

\begin{figure}
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\includegraphics[width=\textwidth]{Figure1.png}
\caption{IS6110 restriction fragment length polymorphism patterns of isolates from patients A, B, and C. Molecular size markers are indicated in lanes 1 and 5.}
\end{figure}
spoligotyping. Spoligotyping is based on the amplification of spacer oligonucleotides between conserved direct-repeat regions. The spoligotype distribution patterns for each of the three patients (Figure 2) confirmed infection with an identical strain of \( M \) \textit{tuberculosis}.

**Discussion**

A child with positive TST results is an indication of recent and possibly ongoing transmission of TB in the community. Finding the source case for the infection is a high priority. A source case investigation or associate investigation is designed to detect the source of transmission in the child's social environment. Unlike adults, children rarely have cavitary lung disease, and small amounts of bacilli are contained in their sputum. In addition, children are less capable of generating forceful coughing, thus limiting an aerosolizing effort of droplet nuclei. As a result, horizontal transmission of \( M \) \textit{tuberculosis} from child to child is rare and the source case is almost always an adult family member.

Most children with TB have no signs or symptoms of classic pulmonary TB disease, and it is only after obtaining positive TST results that a practitioner detects TB transmission. In children with latent TB infection, radiologic analysis of the lungs and pleura are often clinically normal. The impression that "this is just latent tuberculosis" is dangerous, and it is important to recognize that a child with latent tuberculosis is evidence of a contagious tuberculosis case in the community.

The natural history of tuberculosis is a continuum of three stages: exposure, infection, and disease. Tuberculosis prevention is accomplished through disrupting this continuum by discovering and treating the patient, identifying the patient's contacts, and treating those contacts who have latent infection. Conducting an associate investigation will achieve all three objectives. Source cases and other family members infected by these sources are identified through systematic assessment and testing of individuals within the social and physical environments of the infected child and are then treated. Associate and contact investigations are the cornerstone of any TB prevention program. These investigations are designed to identify persons exposed to TB by conducting face-to-face interviews and assessing risk in the home and work environments.

Based on a concentric-circle approach, an investigation prioritizes contacts on the bases of degree of exposure time and individual susceptibility. This prioritization conserves health department resources and focuses the investigation on those who are at greatest risk of becoming infected. In an associate investigation, all adults with significant contact are potential source cases. Once significant exposure to the child has been identified, TST should be performed and chest radiographs should be taken to diagnostically assess for the presence of \( M \) \textit{tuberculosis}. Early detection and treatment of latent TB infections found during an investigation prevents further progression into disease and additional transmission.

If there is evidence of transmission within the family environment, such as persons with positive TST results, abnormal chest radiographs, and/or AFB-positive sputa, the concentric circle is expanded to other contacts in other social environments with similar duration of exposure. Expansion of the investigation ends when the target group infection rate is lower or similar to the level of infection in the community. The investigation process is not complete until a 90-day follow-up skin test is performed. The follow-up TST is necessary to evaluate contacts whose initial skin test was negative, to detect conversion to positive after the time in which the body is developing a cell-mediated immune response to \( M \) \textit{tuberculosis}.

Deoxyribonucleic acid fingerprinting has improved our understanding of the transmission of TB. Molecular epidemiology studies of TB have been used to identify strains of TB involved in laboratory errors and nosocomial transmission. Additionally, DNA fingerprinting has been used successfully to confirm the results of contact investigations and have suggested that a substantial proportion of TB cases in urban areas are due to recent transmission. Detection of same-strain or clustered transmission within epidemiologically identified groups has the potential to recognize outbreaks before they become widespread and aids in program design for prevention and control. In this case, DNA fingerprinting demonstrated that all three of the culture-positive TB cases were infected with the same strain. The arrival of the uncle from Mexico who was infected with the same strain provides evidence that the source case lives in Mexico.

A contact investigation should be conducted whenever a patient is found to have or is suspected of having infectious TB
disease. Evidence of infectious TB disease includes AFB-positive sputa, abnormalities on chest radiographs, and positive TST results, combined with the signs and symptoms of pulmonary infection. An associate investigation should be initiated when a child has positive TST results. Appropriate contact investigations and treatment of TB result in the interruption of transmission and reduction of TB morbidity.

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