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Are Three Sputum Acid-Fast Bacillus Smears Necessary for Discontinuing Tuberculosis Isolation?

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To evaluate the efficacy of three sputum acid-fast bacillus (AFB) smears to rule out pulmonary tuberculosis, sputum AFB smear and culture results were analyzed at two university-affiliated teaching hospitals. The negative predictive value of the smear increased by only 0.2% on days 2 and 3 each, indicating that in low-prevalence populations, there is limited value in requiring three negative sputum AFB smears before discontinuing tuberculosis isolation.

Current guidelines recommend that patients suspected of having active pulmonary tuberculosis (TB) in a health care facility should be placed in a TB isolation room (3). These recommendations state that isolation can be discontinued when the diagnosis of TB is ruled out or when a determination has been made that the patient is noninfectious. Once a patient has been diagnosed with TB, "isolation should be discontinued only when the patient is on effective therapy, is improving clinically, and has had 3 consecutive negative sputum acid fast bacillus (AFB) smear examinations collected on different days" (3).

In clinical practice, patients suspected of having pulmonary TB are placed routinely in TB isolation. Although most United States hospitals require three sputum AFB smears to discontinue isolation, the published literature reveals limited data to support this 3-day requirement (2, 6–8, 10). Therefore, we examined the value of this practice at two New Jersey university-affiliated teaching hospitals.

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Mycobacteriology laboratory records for all sputum AFB smear and culture tests performed at Robert Wood Johnson University Hospital, New Brunswick, N.J., and the Veterans Affairs Medical Center, East Orange, N.J., were reviewed from September 1993 through September 1998.

Specimens from patients who had three separate sputum collections (induced or expectorated) submitted for AFB smear and culture on different days within a 14-day period were included in the analysis. Specimens were not included in the analysis if they were repeat sputum series from a patient already included in the study. Patients who contributed fewer than three sputum AFB specimens within a 14-day period were also excluded.

Sputum smears were decontaminated, digested, and concentrated by standard laboratory methods. Smears were screened by using auramine O stain followed by Kinyoun stain confirmation. Sputum sediments were inoculated to BACTEC 12B broth (Becton Dickinson Microbiology Systems, Sparks, Md.) and nonselective and selective Middlebrook 7H11 solid media. Cultures were incubated at 37°C in a 5% CO_2 incubator for up to 6 weeks. Positive cultures were identified with the AccuProbe DNA hybridization assay (GenProbe, San Diego, Calif.). If more than one specimen was received on a given day, either they were pooled or only the first specimen was included in the data analysis.

The sensitivity, specificity, and negative predictive values (NPVs) of the sputum smear examination were calculated by using the sputum culture results as the "gold standard." Confidence intervals (CI) were calculated by the exact method. For the difference between proportions, the estimations of confidence interval were based on the normal approximation.

During the study period, 5,336 sputum specimens were submitted for AFB smear and culture from 1,981 patients. *M. tuberculosis* grew in 78 specimens from 25 patients at Robert Wood Johnson University Hospital and 125 specimens from 31 patients at Veterans Affairs Medical Center, a total of 203 positive cultures from 56 patients. The overall prevalence of culture-positive *M. tuberculosis* in sputum specimens was 2.83%.

Sputum AFB smears and culture results for *M. tuberculosis* are shown in Table 1. Sputum AFB smears had a sensitivity of 67.5% (95% CI, 60.6 to 73.9) and specificity of 97.5% (95% CI, 97.0 to 97.9) when the data from the two hospitals were combined.

Only 488 patients (24.6%) produced three separate sputum specimens on different days during a 14-day period. Of these, 27 patients had positive cultures. The results of AFB smear and culture categorized by the sequence in which specimens

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TABLE 1. Sputum AFB smear and culture results for *M. tuberculosis*

AFB smear result	No. of samples with *M. tuberculosis* culture result		Total
	Positive	Negative	
Positive	137	130	267
Negative	66	5,003	5,069
Total	203	5,133	5,336

were submitted are shown in Table 2. Eight of 27 (29.6%) patients were AFB smear negative but culture positive for *M. tuberculosis.* Of 19 smear-positive patients, 17 (89.5%) were identified as positive on the first smear, 1 (5.3%) was identified on the second smear, and 1 (5.3%) was identified on the third smear.

The NPV of AFB smears according to the day of the first positive smear is shown in Table 3. As is evident, the incremental value of second and third specimens after a first negative smear was negligible.

In this study the overwhelming majority of patients whose AFB smears were negative and for whom TB isolation was discontinued were identified with the first sputum AFB smear. The incremental benefits of 0.2% with the second specimen and of 0.2% with the third specimen have to be weighed against the costs of additional patient days in TB isolation.

Current recommendations (3) were formulated at a time when there was resurgence in TB (11). However, there is a paucity of published literature directly supporting the current clinical practice of continued TB isolation until three sputum smears are negative.

The 1985 U.S. Public Service Guide for level III laboratories (6) suggested that the number of specimens submitted for culture be determined by the results of early smear examination, and Bates (2) stated that three specimens were sufficient when at least two of the first three smears were positive. Data to support the numbers of smears recommended to diagnose pulmonary TB were derived from field studies in high-prevalence areas in South East Asia or from preselected symptomatic patients with prolonged cough, purulent sputum, and hemoptysis (10). In these populations, two consecutive smears were sufficient for detecting TB.

In a study conducted at the Tuberculosis Chemotherapy Centre, Madras, India, in the 1950s, two immediate (spot)

TABLE 2. Results of AFB smear and culture categorized by the sequence in which specimens were submitted

AFB smear result	No. of patients with *M. tuberculosis* culture result that was:		Total no. of patients
	Positive	Negative	
Positive	19	17	36
First on day 1	17	10	27
First on day 2	1	4	5
First on day 3	1	3	4
Negative	8	444	452
Total	27	461	488

TABLE 3. NPV of AFB smears according to the day of collection

Day	No. of patients with negative result		NPV (%) (95% CI)	Increase (%) in NPV (95% CI)
	Culture	Smear		
1	451	461	97.8 (96.0–99.0)	
2	447	456	98.0 (96.3–99.1)	0.2 (−1.6–2.0)
3	444	452	98.2 (96.5–99.2)	0.2 (−1.6–2.0)

collections were compared with two specimens collected overnight in 348 sputum-positive patients, the great majority of whom had extensive disease (1). For AFB smears, the percentages reported positive were 66.2 and 76.4% for the spot and collection methods, respectively, and 89.7 and 93.7% for culture. Limitations in the data presented make calculation of sensitivity and specificities impossible.

In retrospective studies by MacGregor (7) and Greenbaum et al. (5), it was concluded that no more than three sputum specimens were necessary for the diagnosis of TB if, in fact, the diagnosis was to be made by sputum examination. The same criteria were recommended for discontinuing isolation. However, neither study specifically provided data with regard to the number of smears.

These data have helped form the basis of the current three-specimen requirement for ruling out pulmonary TB. The probable rationale behind this recommendation is that some patients shed mycobacteria irregularly and in small numbers and, thus, increasing the number of specimens would increase the yield (2). With regard to the recommendation for early morning specimens, the assumption is that *M. tuberculosis* would be present in maximum concentration in sputum after pooling overnight in the respiratory tract.

Two recently published reports have provided results similar to the observations in the present study. Nelson et al. (8), in a 10-year retrospective study in Minneapolis, Minn., found that the majority of culture-proven pulmonary TB cases were diagnosed from the first or second sputum specimens and that only rarely was the third specimen of diagnostic value. Craft et al. (4) reviewed 4 years' worth of data at the University of North Carolina and concluded that "modifying the smear policy from three to two negative smears would have resulted in no increased risk of spreading TB and would decrease the number of days patients are unnecessarily placed under airborne precautions" in their institution.

Our study and those cited above raise the question of whether three sputum AFB smears are necessary before discontinuing TB isolation. In the two hospitals that we studied, it took up to 14 days to obtain a third sputum specimen for 488 of 1,981 patients, further pointing to the impracticality of the recommendations in routine clinical practice. Whether multiple negative sputum AFB smears obtained during a shorter period such as 24 h would obviate the collections on subsequent days has not been studied.

Since the incidence of TB is once again declining in the United States (9), a reassessment of the requirement for three negative sputum smears to discontinue TB isolation seems warranted. However, once the initial AFB smear, or arguably two smears, is negative, the additional specimens for culture

can be obtained after isolation is discontinued. Exceptions to this recommendation may be necessary when the pretest probability of TB is high.

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