Microscopy — misc

Evaluation of the Acid-Fast Smear

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A retrospective analysis of the data involving all specimens submitted to the mycobacteriology laboratory over the past 20 months supports the continued use of the acid-fast smear as an aid in the detection of tuberculosis.

The microscopic detection of acid-fast bacilli by auramine-rhodamine or Ziehl-Neelsen staining has been used as an aid in tuberculosis diagnosis for many years. Two recent reports, however, have indicated that acid-fast staining as a screening technique for the detection of tuberculosis is unreliable in populations, such as in the United States, which have a low incidence of tuberculosis. Boyd and Marr (1) reported that only 22% (26 out of 118) of all positive cultures were also smear positive (true positive), and the rate reported by Marraro et al. (2) was 24.1% (14 out of 58). Boyd and Marr (1) further determined that 99.3% (4,420 out of 4,452) of all cultures that were negative for growth also showed no microscopic evidence of acid-fast bacilli and were classified as true negative. Marraro's true negative rate was 99.5% (1,915 out of 1,925). These papers prompted us to review our records of the past few years and analyze our data with regard to paired acid-fast smears and cultures to determine the reliability of this procedure in our laboratory.

All specimens for the year 1974 and through August 1975 were included in this study, with the exception of those that showed contamination upon culture; the records of 6,199 specimens on which both smear and culture were performed were used to compile the data. Specimens were digested and concentrated by the acetyl-cysteine-alkali procedure (4), inoculated onto Lowenstein-Jensen slants, incubated at 37°C in 5% CO_2, and observed for growth over an 8-week period. Smears were stained by the Ziehl-Neelsen method (5) or an auramine-rhodamine technique (3).

To compare our results with Boyd and Marr (1) and Marraro et al. (2), we categorized our smears according to their criteria: (i) a true positive smear showed the presence of acid-fast bacilli and culture of the material yielded mycobacteria; (ii) a true negative smear was one in which no organisms were seen on smear or grown in culture; (iii) a false-positive smear showed the presence of acid-fast bacilli, but culture of the material produced no growth; (iv) a false-negative smear showed no microscopic evidence of acid-fast bacilli, but culture of the material produced growth of mycobacteria.

Table 1 shows the results obtained in our laboratory when smears were compared with cultures. Of the 227 culture-positive specimens, 97 were detected by acid-fast staining, and of the 5,972 culture-negative specimens, 7 acid-fast smears were falsely positive. It is readily apparent that there are numerous ways of determining percentages from Table 1. To compare our results with those of the other two laboratories, all data were calculated based on the above criteria. Table 2 shows that although the true negative rate (specificity) of the acid-fast smear is very high (greater than 99%) in all three laboratories, in our laboratory the specificity was significantly greater ($P < 0.001$). These data suggest that false-positive results are comparatively rare, and therefore a positive smear can be relied upon as a good diagnostic indicator, especially when associated with significant clinical manifestations.

Although the sensitivity or true positive rate of the test in our laboratory (42.7%) was not as good as one would like, it was significantly greater ($P < 0.001$) than that of the other two laboratories (22 and 24.1%). From a practical point of view, this means that approximately 50% of our positive cultures were initially detected by smear, whereas approximately 25% of positive cultures were smear detected in the other two laboratories. Differences in tuberculosis incidence throughout the United States

TABLE 1. *Comparison of acid-fast smears with cultures*

Culture	Smear		Total
	Positive	Negative	
Positive	97	130	227
Negative	7	5,965	5,972
Total	104	6,095	6,199

TABLE 2. *Relative paired-sample analyses of acid-fast smears and cultures*

Results	Boyd and Marr (1)	Marraro et al. (2)	Burdash et al.[a]
True positive (sensitivity)	26/118 (22%)	14/58 (24.1%)	97/227 (42.7%)
True negative (specificity)	4,420/4,452 (99.3%)	1,915/1,925 (99.5%)	5,965/5,972 (99.9%)
False positive	32/4,452 (0.7%)	10/1925 (0.5%)	7/5,972 (0.1%)
False negative	92/118 (78%)	44/58 (75.9%)	130/227 (57.3%)

[a] All values are significantly different from those of Boyd and Marr and Marraro et al. ($P < 0.001$).

should not account for such wide variation of the data nor should differences in staining by auramine-rhodamine or Ziehl-Neelsen techniques. At this point, we are unable to explain these differences since all three studies were done in reputable laboratories in virtually the same manner using all types of patients and specimens. We believe our data support the continued use of the acid-fast smear as an aid in the diagnosis of mycobacterial infections. Of the seven smears that were falsely positive, five were from patients who had other positive smears and cultures indicating mycobacterial infection, and a sixth was from a patient undergoing prolonged anti-tuberculosis chemotherapy. The fact that the smear examination detected almost half of all specimens that eventually grew out mycobacteria coupled with the high specificity of the test indicates that in our laboratory the positive acid-fast smear is a useful aid in alerting the clinician to the possibility of tuberculosis in his patient.

LITERATURE CITED

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