

Predictive Value of the Acid Fast Smear for Detection of *Mycobacterium tuberculosis* in Respiratory Specimens in a Reference Center of HIV/Aids in Rio de Janeiro, Brazil

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In order to evaluate the predictive value of acid fast bacilli (AFB) smear for the diagnosis of Mycobacterium tuberculosis in respiratory specimens in a setting with a high prevalence of Aids and an unknown prevalence of nontuberculous mycobacteria (NTM), we retrospectively examined specimens cultured for mycobacteria between 1 September 1993 and 30 September 1994 and medical records of patients with positive culture in a General Hospital, Aids reference in Rio de Janeiro, Brazil. Seventy three per cent (1517/2077) of samples were respiratory specimens and mycobacteria were recovered from 20.6% (313/1517) of these. M. tuberculosis was identified in 94.2% (295/313) and NTM in 5.8% (18/313). The yield of positive AFB smear and of positive culture was 6.1% (93/1517) and 20.6% (313/1517), respectively. The positive predictive value (PPV) of AFB for M. tuberculosis was 98.4% in expectorated sputum and 96.4% in bronchoalveolar lavage. Forty four percent (130/295) of specimens with positive culture for M. tuberculosis and 66.7% (12/18) for NTM were from patients HIV positive. The conclusion was that in our study population, the PPV of AFB for M. tuberculosis in respiratory specimens was high and the prevalence of NTM was low despite the high prevalence of HIV positive.

Key words: *Mycobacterium tuberculosis* - acid fast smear - non tuberculous mycobacteria - HIV/Aids

Before the acquired immunodeficiency syndrome (Aids) epidemic, infections with *Mycobacterium tuberculosis* (*M.tb*) and nontuberculous mycobacteria (NTM) were recognized as an uncommon cause of disease in developed countries (Horsburgh Jr 1991). Where infection with the HIV became prevalent, the incidence of mycobacteria increased (Horsburgh Jr 1991, Kritski et al. 1995, Raviglione et al. 1995). Although the impact of HIV infection on the incidence of *M.tb* has been well described in developing countries, its impact on the incidence and prevalence of NTM has not (Barreto et al. 1993, Raviglione et al. 1995, Shafer & Edlin 1996). Detection of acid fast bacilli (AFB) in respiratory specimens is the diagnostic approach for the diagnosis of pulmonary tuberculosis (TB),

as recommended by the World Health Organization (WHO 1993). Since NTM are indistinguishable from *M.tb* in AFB smear, the value of positive AFB in respiratory specimens for predicting pulmonary TB is unclear, especially where the prevalence of NTM is unknown. In Rio de Janeiro, Brazil, the prevalence of co-infection of HIV and disseminated mycobacterial disease is expected to be high (Grinsztejn et al. 1997, Kritski et al. 1998). We therefore conducted a study in a General Hospital in Rio de Janeiro with a high prevalence of HIV and mycobacterial infections. We retrospectively examined specimens cultured for mycobacteria and medical records of cases with positive culture for mycobacterium and the predictive value of AFB smear for *M.tb* was determined.

MATERIALS AND METHODS

The medical records of patients from whom clinical specimens were culture positive for mycobacteria were reviewed. The clinical specimens were analyzed by the Bacteriology Laboratory of the Clementino Fraga Filho Hospital of Rio de Janeiro Federal University, an Aids Reference Center. All materials were collected, cultured and smeared for AFB between 1 September 1993 and 30 September

This study was supported by International Aids training grant # D43-TW00003 from Fogarty International Center, NIH, USA.

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Received 13 January 1999

Accepted 7 July 1999

1994. During this period there were no changes in the laboratory methods used for the culture of mycobacteria. The laboratory methods for processing specimens, for cultures and for AFB smears followed standard methods (Kent & Kubica 1985). The respiratory specimens were decontaminated with 4% NaOH. All fluids except blood were plated onto Löwestein Jensen medium and smeared for AFB with the Ziehl Neelsen technique after sedimentation at 3000 x g. For blood specimens, only culture in Löwestein Jensen medium was performed. Cultures were reviewed weekly for growth, and those cultures showing no growth were incubated for eight weeks before they were discarded. All specimens that were culture positive for mycobacteria were tested to distinguish *M.tb* from NTM and organisms belonging to *M. avium* complex (MAC) were identified. HIV antibody counseling and testing was offered to those patients with clinical symptoms or risk factors for Aids. The serum sample was initially screened for HIV antibody by enzyme-linked immunosorbent assay (Orth Diagnostic Systems, Raritan, NJ, USA). Repeatedly reactive samples were submitted to Western blot analysis. Interpretation of Western blot results followed the manufacturer's instruction.

RESULTS

Mycobacteria was recovered from 17.5% (363/2077) of all specimens cultured. We excluded 38 AFB smear positive specimens which were either culture negative (n = 27) or contaminated (n = 11). Among those specimens that were culture-positive, *M.tb* was identified in 93.1% (338/363) and NTM in 6.9% (25/363). The 338 specimens of *M.tb* refer to 294 patients and the 25 specimens of NTM to 20 patients. Seventy three per cent (1517/2077) of samples cultured were respiratory specimens.

Mycobacteria was recovered from 20.6% (313/1517) respiratory specimens, with 65.4% (205/313) in expectorated sputum, 32.6% (102/313) in bronchoalveolar lavage (BAL) and 2% (6/313) in bronchial aspirate. *M.tb* was identified in 94.2% (295/313) and NTM in 5.8% (18/313). *M.tb* was isolated in 93.6% (192/205) of expectorated sputum and in 95% (97/102) of BAL while NTM was isolated in 6.4% (13/205) and 5% (5/102), respectively (Table). Among NTM, 27% (5/18) were identified as *avium* complex and the others were not identified. The rate of smear positivity calculated for expectorated sputum was 32.2% (62/192) for specimens with *M.tb* and 7.7% (1/13) in specimens with NTM (Table). The rate of AFB positivity for *M.tb* in expectored sputum from HIV positive patients was 29.7% (25/84) and in patients with HIV negative or without HIV tested was 34.2% (37/108). The rates of smear positivity for BAL were 27.8% (27/97) for *M.tb* and 20% (1/5) for NTM (Table). The rate of AFB positivity in BAL from HIV positive patients was 23.8% (10/42) and in patients with HIV negative or without HIV tested was 30.9% (17/55). The positive predictive value (PPV) of AFB for *M.tb* in all cultures positive for mycobacteria was 98.4% (62/63) in expectorated sputum and 96.4% (27/28) in BAL. The yield of positive AFB during the period of the study was 6.6% (63/952) for sputum specimens and 5.3% (27/505) for BAL specimens while the yield of positive culture was 21.5% (205/952) and 19.2% (97/505), respectively.

Forty four percent (130/294) of patients with positive culture for *M.tb* and 60% (12/20) with NTM were HIV positive. Among the 18 patients that have respiratory specimens culture-positive for NTM, 66.6% (12/18) were male, with an age range between 25 and 53 years (median 37.1). In 16.6% (3/18) of these patients, disseminated infection were con-

TABLE

Results of acid fast bacilli (AFB) smear of 313 respiratory specimens with positive culture for mycobacteria, 1993-1994

Specimen	<i>Mycobacterium tuberculosis (M.tb)</i>		Nontuberculous mycobacteria (NTM)		Total of positive cultures
	Positive culture for <i>M.tb</i>	AFB+	Positive culture for NTM	AFB+	
	N (HIV+)	N (HIV +)	N (HIV+)	N (HIV+)	
Sputum (n = 952)	192 (84)	62 (25)	13 (9)	1 (1)	205
Bronchoalveolar lavage (n = 505)	97 (42)	27(10)	5 (3)	1 (1)	102
Bronchial aspirate (n = 60)	6 (2)	2 (1)	0	0	6

firmed as well in the two remaining patients with NTM infection (four blood specimens and one bone marrow specimen). The five cases with disseminated disease were HIV positive.

DISCUSSION

In this study, the prevalence of 6.9% culture positive specimens for NTM was lower than that described in the United States and Europe (Good & Snider 1983, Horsburgh Jr 1991, WHO 1993, Yajko et al. 1994). Yajko et al. (1994) reported 51% NTM among 2139 mycobacteria cultured in San Francisco. This figure was higher than the 35% cases of NTM found in 1983, prior to the HIV epidemic, in all mycobacteria isolated in the United States (Good & Snider 1983).

In São Paulo, Brazil, 0.87% (23/2628) of Aids patients had disseminated disease due infection with NTM in a study performed from 1990-1992 (Barreto et al. 1992). In Rio de Janeiro, bacillemia was detected in 30 of 42 patients with *Mycobacterium* isolated (Grinztejn et al. 1997). MAC was detected in 11 (28.2%) and *M.tb* in 19 (71.8%). Unfortunately, in those two prospective studies no data regarding the yield of AFB and culture in respiratory specimens were showed. In our retrospective study, the occurrence of disseminated mycobacterial disease was lower because we focused mainly on analyses respiratory specimens.

Before the Aids epidemic, 40 to 70% of patients with *M.tb* isolated in culture had positive AFB smears (Toman 1979). Among patients with Aids, these figures tend to be lower (WHO 1993, Shafer & Edlin 1996). In our General Hospital, an Aids reference center, the smear positivity among the culture positive specimens for *M.tb* was lower in expectorated sputum (32%) and in bronchoalveolar lavage (27%). Among all specimens of sputum evaluated, the yield of AFB in the sputum was 6.6% and of culture for *M.tb*, 21.5%. These data suggest that culture is indicated for diagnosis of TB in these patients, mainly in a hospital with a high prevalence of co-infection TB/HIV. These results are different from those found by Elliot et al. (1990) in Zambia, where the yield of AFB and culture was quite similar to that of clinical diagnosis of tuberculosis.

Although Klein et al. (1989) has reported a smear-positivity rate of sputum specimens from HIV positive patients lower than the rate from individuals not infected with HIV, Yajko et al. (1994) and Theuer et al. (1990) found no difference between smear-positivity rate of sputum specimens of these two groups of patients. In our study, the smear positivity rate of sputum and BAL specimens from HIV positive patients tend to be lower than that observed among specimens from patients HIV negative and those with no HIV test performed.

We found AFB positive in 11% (2/18) of respiratory specimens which were culture positive for NTM, even with the high prevalence of cases with positive HIV. The prevalence of AFB positivity among NTM seems to vary according to the region studied. In the United States, 6% (15/232) of the expectorated sputum specimens that were culture positive for MAC were AFB positive (Yajko et al. 1994). In Hong Kong, 86% of sputum specimens from 28 patients with progressive pulmonary disease due to NTM had AFB positive smears, whereas only 2% of 134 patients with transient isolation of NTM or without evidence of progressive pulmonary disease (contaminated sputum specimens) had AFB positive smears (Hosker et al. 1995).

Yajko et al. (1994) reported that the PPV of the AFB smear for *M.tb* was high (92%) for sputum and relatively low (71%) for BAL specimens. In our study, the PPV of the AFB for *M.tb* in expectorated sputum (98.4%) and in BAL specimens (96.4%) remained as high as the level reported before the Aids epidemic (82-100%) (Strumph et al. 1979, Murray et al. 1980).

The data presented in this study demonstrate that in a setting with a high prevalence of Aids, the PPV of AFB for *M.tb* in respiratory specimens from expectorated sputum or BAL is high and the prevalence of NTM is low. Our data also demonstrate the high sensitivity show the importance in these specimens of culture for the tuberculosis diagnosis in this setting.

ACKNOWLEDGMENT

To Dr Adalbert Laszlo (National Reference Centre for Tuberculosis, Ottawa, Canada), for his revision of the text and suggestions.

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