Mycobacterium gordonae Pseudoinfection Associated with a Contaminated Antimicrobial Solution

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At Yale-New Haven Hospital, 46 specimens submitted for mycobacterial culture during an 8-week period in 1989 were positive for *Mycobacterium gordonae*, a nontuberculous acid-fast bacterium (AFB) of low pathogenicity. The specimens were submitted from 34 patients who came from various inpatient and outpatient services. Four patients were begun on antimycobacterial therapy on the basis of an AFB isolate which was later identified as *M. gordonae*. Isolation of *M. gordonae* was associated with use of the BACTEC TB system (BACTEC TB; Becton Dickinson Diagnostic Instrument Systems, Towson, Md.) and an antimicrobial solution, BACTEC PANTA PLUS (PANTA; Becton Dickinson Diagnostic Instrument Systems). The manufacturer reported that two lots (B9K1 and C9K1) of PANTA kits containing a single production lot (N8C1) of PANTA, which had been shipped to 173 laboratories, had been contaminated with *M. gordonae*. A survey of mycobacteriology laboratories in the United States revealed that, during April to July 1989, the *M. gordonae* isolation rate was 5.8/1,000 AFB specimens processed at laboratories that did not use BACTEC TB, 11.4/1,000 AFB specimens at laboratories that used BACTEC TB but not the implicated lot of PANTA, and 23.5/1,000 AFB specimens at laboratories that used BACTEC TB and the lot of implicated PANTA. Intrinsic contamination of PANTA was attributed to ineffective sterilization of water used in the manufacturing process and was not detected prior to product shipment because cultures for AFB were not part of the quality control regimen. This episode emphasizes that clinical laboratories can detect pseudoepidemics promptly if they are alert to abrupt increases in isolation rates, especially of unusual or generally nonpathogenic organisms.

Mycobacterium gordonae, a nontuberculous mycobacterium, is commonly found in water and sputum (22). In a survey of state health departments, it was the third most commonly isolated acid-fast bacterium (AFB), comprising 14.6% of all isolates (7). Despite this ubiquity, *M. gordonae* rarely causes disease, and the vast majority of isolates represent harmless commensal or contaminant organisms introduced during specimen collection or processing (22). *M. gordonae* isolates occurring in the absence of invasive disease may create diagnostic confusion and may prompt unnecessary antimycobacterial therapy, especially in the period between detection of AFB growth in laboratory cultures and identification of the growth as *M. gordonae*.

The BACTEC TB system (BACTEC TB; Becton Dickinson Diagnostic Instrument Systems, Towson, Md.) is a radiometric method for detection of mycobacterial growth in vials of medium containing ^{14}C-labeled substrate (BACTEC 12B medium). A solution of five antimicrobial agents (polymyxin B, amphotericin B, nalidixic acid, trimethoprim, and azlocillin), BACTEC PANTA PLUS (PANTA; Becton Dickinson Diagnostic Instruments), is used to suppress growth of non-AFB microorganisms (19). BACTEC TB allows detection of AFB more rapidly than conventional solid AFB medium does (17). We present here the results of our investigation of BACTEC TB cultures contaminated with *M. gordonae*. To our knowledge, this is the first published report of an episode of AFB pseudoinfection caused by a commercially distributed product.

MATERIALS AND METHODS

Background. Yale-New Haven Hospital (Y-NHH) is an 818-bed acute- and tertiary-care facility. During May and June 1989, the Clinical Microbiology Laboratory at Y-NHH noted an unusually large number of *M. gordonae* isolates. Despite inoculation of most specimens into a number of AFB media, most *M. gordonae* isolates grew only in BACTEC TB, suggesting contamination of cultures.

Investigation at Y-NHH. Laboratory records for the period 1 November 1985 to 1 August 1989 were reviewed for the numbers of AFB specimens processed and *M. gordonae* recovered. Based on these results, a case-specimen was defined as a specimen submitted from 4 May to 1 July 1989 from which *M. gordonae* was isolated only in BACTEC TB. The specimen site, requesting service, and, when known, use of PANTA was recorded for case-specimens. A case-patient was defined as any patient from whom a case-specimen was submitted. Inpatient medical records of case-patients were reviewed. Laboratory procedures were reviewed, and technicians who processed AFB specimens were observed.

Information from the manufacturer. Information regarding the manufacturer's internal investigation and response to the incident was supplied by Becton Dickinson Diagnostic Instrument Systems.

Survey of mycobacteriology laboratories. In January 1990, we conducted a written survey of 607 mycobacteriology laboratories regarding (i) the numbers of AFB specimens processed and the numbers of *M. gordonae* isolated from all media types from 1 April 1989 to 1 August 1989; (ii) whether the laboratory used BACTEC TB during this period and, if so, whether a letter from the manufacturer recalling PANTA
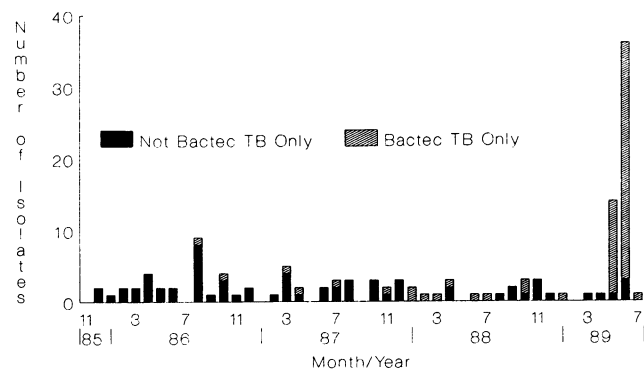
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FIG. 1. *M. gordonae* isolates by date and culture medium, isolated at Y-NHH from November 1985 to July 1989.

TABLE 1. *M. gordonae* isolation rates in the base-line and epidemic periods by type of media at Y-NHH from 1 November 1985 to 1 July 1989

Medium	Base-line period ($n = 12,058^a$)		Epidemic period ($n = 603^a$)		P value
	No. of isolates	Rate[b]	No. of isolates	Rate[b]	
Bactec TB only	16	1.3	46	76.3	<0.0001
Bactec TB and other	17	1.4	3	5.0	0.067
Other only	45	3.7	1	1.7	0.725
Unknown	0	0	1	1.7	
Total	78	6.5	51	84.6	<0.0001

[a] Number of specimens processed for AFB.
[b] Rate per 1,000 specimens processed for AFB.

lot N8C1 was received; (iii) the type and numbers of other culture media used; and (iv) the pretreatment regimen for specimens from nonsterile sites to minimize growth of non-AFB organisms (decontamination). Laboratories responding to the survey were classified as not using BACTEC TB, using BACTEC TB but not receiving PANTA lot N8C1, or using BACTEC TB and receiving PANTA lot N8C1 (defined as those that reported receiving a letter from the manufacturer recalling this lot).

Data analysis. Data were analyzed by using Epiinfo Software (Centers for Disease Control, Atlanta, Ga.). Proportions were compared by using the Yates corrected chi-square test. Survey data were used to calculate *M. gordonae* isolation rates per 1,000 specimens processed for AFB at individual laboratories. Stratified relative risks were determined by the Mantel-Haenszel procedure. Pooled isolation rates for groups of laboratories were calculated from combined numbers of *M. gordonae* isolates and specimens processed for AFB.

RESULTS

Investigation at Y-NHH. When the numbers of *M. gordonae* isolated at Y-NHH between 1 November 1985 and 1 August 1989 were examined, an abrupt increase in isolates was seen in May and June 1989 (Fig. 1). There was a highly significant increase in the *M. gordonae* isolation rate during the "epidemic" period (4 May 1989 to 1 July 1989) compared with that during the base-line period (1 November 1985 to 1 May 1989; Table 1). When isolation rates by medium type were compared during these periods, the numbers of *M. gordonae* isolated only from BACTEC TB increased significantly from 16/12,056 to 46/603 ($P < 0.001$), whereas rates of isolation from other media or simultaneously from BACTEC TB and other media showed nonsignificant changes (Table 1). These data confirm that an outbreak of *M. gordonae* isolates occurred and was associated with the use of the BACTEC TB system.

The 46 specimens that met the case definition came from a variety of sites: 26 (56.5%) from sputum; 7 (15.2%) from urine; 4 (8.7%) from cerebrospinal fluid (CSF); 4 (8.7%) from tissue or wound; and 1 (2.2%) each from blood, bronchial brushing, bronchial washing, nasal swab, and a gastric aspirate. The case-specimens were submitted from several inpatient services and outpatient areas.

Smears of all case-specimens were initially negative for AFB. AFB were isolated a median of 21 days after specimen submission and were identified as nontuberculous mycobac-

teria by using BACTEC TB a median of 5 days after isolation. Final identification as *M. gordonae* was completed a median of 37 days after specimen submission. Prior to suspicion in mid-June 1989 that the isolates represented contamination, attending physicians were notified as soon as growth of AFB was detected; subsequently, AFB isolated only from BACTEC TB were not reported until identification had been completed, and reports of *M. gordonae* isolation included a statement that contamination was suspected.

The 46 case-specimens came from 34 case-patients with a median age of 55 years (range, 1 to 82 years). Nine case-patients (26.5%) had human immunodeficiency virus infection, and five (14.7%) had died as of 10 October 1989. Five case-patients (14.7%) were on antimycobacterial therapy at the time of specimen submission; of 29 patients who were not already on antimycobacterial therapy, four (13.8%) were begun on therapy on the basis of a culture positive for AFB which later proved to be *M. gordonae*.

A review of laboratory procedures revealed that, other than for specimen inoculation, BACTEC TB vials were entered only to inoculate the antimicrobial supplement PANTA. Observations of the technicians who processed the specimens did not identify any breaks in aseptic technique.

Specimens from normally sterile sites such as blood were inoculated in parallel into BACTEC TB vials with and without PANTA, affording an opportunity to assess the possibility of contamination of this antimicrobial solution. Since use of PANTA was recorded only on the BACTEC TB vials themselves, information on the use of PANTA was available only for vials which were not discarded. Sixty-five BACTEC TB vials positive for an AFB during the epidemic period, which had been set aside for use in an unrelated project, were available for evaluation of possible contamination of PANTA. *M. gordonae* was isolated significantly more often from BACTEC TB vials to which PANTA had been added than from vials inoculated without PANTA (32 of 57 vials versus 0 of 8 vials; $P < 0.005$), suggesting that PANTA was associated with *M. gordonae* pseudoinfection. Because of the delay from the time of the contamination to the investigation, we were unable to obtain any vials of the implicated lot of PANTA for culture.

Information from the manufacturer of BACTEC TB. The manufacturer of BACTEC TB reported that 20 other laboratories had reported increased numbers of *M. gordonae* isolates, and all but 1 had received PANTA kits from lot B9K1, lot C9K1, or both, which were prepared from a single PANTA production lot (N8C1). Cultures of six of seven vials

TABLE 2. Laboratory characteristics and pooled *M. gordonae* isolation rates by use of the BACTEC TB system and receipt of PANTA (lot N8C1) from 1 April to 1 October 1989

Laboratory characteristics	No. of AFB media used (mean)	No. of specimens processed for AFB		*M. gordonae*	
		Per laboratory mean	Total	No. isolated	Isolation rate/1,000[a]
BACTEC TB nonusers (*n* = 202)	2.3	583	100,256	583	5.8
BACTEC TB users that:					
Did not receive PANTA lot N8C1 (*n* = 35)	2.6	868	30,372	347	11.5
Received PANTA lot N8C1 (*n* = 32)	2.7	1,137	36,375	854	23.5

[a] Pooled *M. gordonae* isolation rate per 1,000 specimens processed for AFB.

of PANTA from this lot performed by the manufacturer were reported to be positive for *M. gordonae*. Cultures were positive regardless of whether the dry PANTA was dissolved in sterile water or in the diluent supplied in PANTA kits, indicating contamination of the antibiotic powder itself. Cultures of several other lots of PANTA and cultures of BACTEC TB media were reported to be negative. According to the manufacturer, recall letters dated 8 August 1989 were sent to 173 laboratories that had received this lot of PANTA.

The manufacturer's review did not reveal a definite mechanism of contamination, but suggested that the deionized water used in preparing one batch of PANTA had not been effectively autoclaved. PANTA is not terminally sterilized after production because of fears that heat would inactivate the antimicrobial agents and that filtration would reduce the concentration of amphotericin B. Before the episode of contamination, quality control cultures for aerobic and anaerobic bacteria, but not AFB, were performed. Subsequently, cultures for AFB were added.

Results of survey of mycobacteriology laboratories. The survey of mycobacteriology laboratories was returned by 321 (53%) laboratories. Laboratories were excluded from analysis if they did not process AFB specimens (*n* = 7), did not provide numbers of total specimens processed and numbers of *M. gordonae* isolated (*n* = 38), could not be classified as either using or not using BACTEC TB on a routine basis (*n* = 5), or were not located in the United States (*n* = 2). After these exclusions, data from 269 laboratories were analyzed.

The pooled *M. gordonae* isolation rate was lowest for laboratories that did not use BACTEC TB (5.8/1,000), somewhat higher for those that used BACTEC TB but that did not receive PANTA lot N8C1 (11.4/1,000), and highest for those that used BACTEC TB and that received PANTA lot N8C1 (23.5/1,000; Table 2). The median duration of specimen decontamination was 15 min for all three groups and mean numbers of AFB media used were similar, but laboratories that used BACTEC TB tended to process more specimens than laboratories that did not use BACTEC TB (Table 2).

M. gordonae isolation rates at individual laboratories are shown in Fig. 2. The 32 laboratories that used BACTEC TB and that received PANTA lot N8C1 had the highest isolation rates. Within this group, however, there was considerable variation: 15 (47%) laboratories had rates below 10/1,000, and 11 (34%) laboratories had rates above 30/1,000.

Because of differences between BACTEC TB users and nonusers in the numbers of specimens processed, we calculated Mantel-Haenszel relative risks, stratifying laboratories by numbers of specimens processed per 4 months (<200 specimens [*n* = 91], 200 to 500 specimens [*n* = 89], and >500 specimens [*n* = 89]). Compared with laboratories that did

not use BACTEC TB, laboratories that used BACTEC TB and that received PANTA lot N8C1 were significantly more likely to have had an *M. gordonae* isolation rate of $\geq 20/1,000$ (relative risk = 5.5; 95% confidence interval = 2.6, 11.6; $P < 0.001$). There was no significant difference between BACTEC TB users that did not receive PANTA lot N8C1 and BACTEC TB nonusers (relative risk = 2.4, 95% confidence interval = 0.9, 6.7; $P = 0.16$).

DISCUSSION

There are a number of published case reports of invasive disease caused by *M. gordonae*. Because of the frequency of contamination of cultures with this common environmental organism, cases in which *M. gordonae* has been isolated only once may represent contamination rather than true infection (2, 9, 13, 18). In the absence of clinical evidence of infection, colonization of the respiratory tract may explain some sputum isolates (11). Reported cases in which *M. gordonae* was isolated at least twice, and in some cases multiple times, include three pulmonary infections (4, 10, 12), a peritoneal infection in a patient receiving continuous ambulatory peritoneal dialysis (15), meningitis in a hydrocephalic child with a ventriculoatrial shunt (6), endocarditis of a prosthetic valve (14), cutaneous infections of the foot and wrist (16), and a patient with the acquired immune deficiency syndrome with *M. gordonae* isolated from both sputum and bone marrow (3). These case reports indicate that, since *M. gordonae* occasionally causes disease, isolates cannot always be attributed to contamination or colonization. As expected of an organism of low pathogenicity, no outbreak or cluster of true *M. gordonae* infections has been reported.
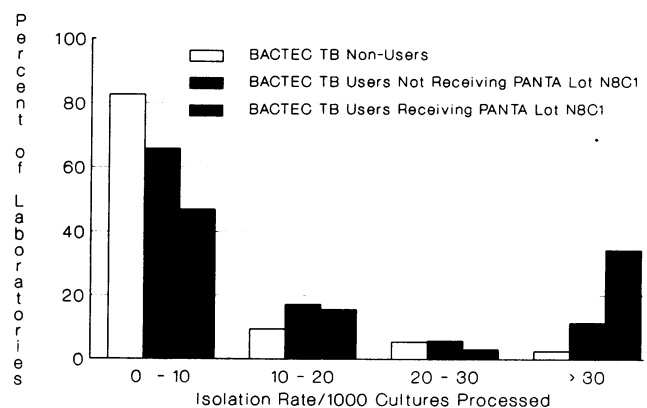

FIG. 2. Distribution of U.S. laboratory *M. gordonae* isolation rates by BACTEC TB system use and receipt of PANTA lot N8C1 from 1 April to 1 August 1989.

M. gordonae has, however, been implicated in a number of pseudoepidemics. Nonsterile deionized water in a pathology laboratory was the apparent cause of a cluster of spurious *M. gordonae* isolates (21). *M. gordonae* isolation from sputum specimens has been attributed to rinsing of patients' mouths with tap water prior to sputum induction with aerosol (5). *M. gordonae* was isolated from bronchoscopy specimens of 52 individuals at Y-NHH from January 1975 through June 1977. The contamination was attributed to green dye added to topical anesthetic by one physician. The low pathogenic potential of *M. gordonae* was illustrated by the absence of clinical infections attributed to this organism, despite inoculation of an estimated 500,000 organisms into each patient's respiratory tract (20). To our knowledge, there have been no previous published reports of pseudoinfection with *M. gordonae* (or any other AFB) caused by contamination of a commercially distributed product.

Problems which may be caused by episodes of pseudoinfection include delay of appropriate therapy, overgrowth of cultures with contaminants, and risks and costs associated with further diagnostic procedures and unnecessary therapy (8). The clinical laboratory must expend time and resources to identify the spurious isolates, determine their source, and notify physicians of the problem. Pseudoinfection with mycobacteria may be particularly troublesome because of the long periods needed for growth and identification of isolates.

Our investigation at Y-NHH pointed to pseudoinfection since case-specimens came from a variety of sites and services and isolation of *M. gordonae* was associated with use of BACTEC TB and PANTA. A median period of 37 days elapsed between detection of mycobacterial growth and identification of the growth as *M. gordonae*. During this period, four patients were started on antimycobacterial therapy. The potential for adverse effects from unnecessary antimycobacterial therapy (1) clearly existed. The number of patients receiving such unnecessary therapy was probably limited by early recognition of the problem by laboratory personnel.

The results of our survey of mycobacteriology laboratories must be interpreted with caution because of the relatively low response rate (53%), the small number of responding laboratories that received the implicated PANTA (n = 32), and the possibility that laboratories with higher *M. gordonae* isolation rates may have been more likely to return the questionnaire. Nevertheless, the survey provides evidence that, during the period that lot N8C1 was reported to have been in use, laboratories that received PANTA lot N8C1 had higher *M. gordonae* isolation rates than did laboratories that did not use BACTEC TB. Laboratories that used BACTEC TB but not the implicated PANTA also had higher isolation rates than did BACTEC TB nonusers, although the difference was not statistically significant. The higher rate among BACTEC TB users that did not receive the implicated PANTA may have been due to greater recovery of naturally occurring *M. gordonae* by BACTEC TB.

The survey also revealed that only some of the laboratories that used BACTEC TB and that received PANTA lot N8C1 were significantly affected by this episode of contamination. Such selective involvement may have been caused by differences in laboratory procedures not covered by our survey or may indicate that not all vials of PANTA lot N8C1 were contaminated. Also, the dates covered by our questionnaire (1 April to 1 August 1989) may not have corresponded with the dates of use of the implicated lot of PANTA, the peak *M. gordonae* isolation at all laboratories, or both.

The 202 laboratories in our survey that did not use BACTEC TB reported 583 *M. gordonae* isolates over the 4-month period. Based on what is known about this organism from published literature, we can assume that few if any of the patients from whom these isolates were recovered had clinical infections with *M. gordonae*. These isolates therefore created the potential for clinical confusion and unnecessary antimycobacterial therapy. Five (2.5%) laboratories that did not use BACTEC TB had *M. gordonae* isolation rates above 30/1,000 over the 4-month period. These laboratories may have lower isolation rates when measured over longer time periods. However, laboratories with continued high *M. gordonae* isolation rates should review specimen collection and processing procedures to identify possible sources of contamination.

Appropriate quality control should prevent the distribution of contaminated products. Before this episode, the manufacturer's quality control procedures for PANTA did not include culturing for AFB; following this episode, the manufacturer has reported that culturing for AFB has been added. This episode highlights the fact that clinical laboratories must critically evaluate increases in isolation rates, especially when unusual or generally nonpathogenic organisms are involved, so that pseudoepidemics can be detected promptly.

ACKNOWLEDGMENTS

LITERATURE CITED

1. **Alford, R. H.** 1990. Antimycobacterial agents, p. 350–360. *In* G. L. Mandell, R. G. Douglas, and J. E. Bennett (ed.), Principles and practice of infectious diseases, 3rd ed. Churchill Livingstone, New York.
2. **Berman, L. B.** 1983. Infection of synovial tissue by *Mycobacterium gordonae*. Can. Med. Assoc. J. **129:**1078–1079.
3. **Chan, J., J. C. McKitrick, and R. S. Klein.** 1984. *Mycobacterium gordonae* in the acquired immune deficiency syndrome. Ann. Intern. Med. **101:**400.
4. **Douglas, J. D., M. A. Calder, Y. F. J. Choo-Kang, and A. G. Leitch.** 1986. *Mycobacterium gordonae*: a new pathogen? Thorax **41:**152–153.
5. **Gangadharem, P. R. J., J. A. Lockhart, R. J. Awe, and D. E. Jenkins.** 1976. Mycobacterial contamination through tap water. Am. Rev. Respir. Dis. **113:**894.
6. **Gonzales, E. P., R. M. N. Crosby, and S. H. Walker.** 1971. *Mycobacterium aquae* infection in a hydrocephalic child (*Mycobacterium aquae* meningitis). Pediatrics **48:**974–977.
7. **Good, R. C., and D. Snider.** 1982. Isolation of nontuberculous mycobacteria in the United States, 1980. J. Infect. Dis. **146:**829–833.
8. **Goodman, R. A., J. D. Smith, G. P. Kubica, E. M. Dougherty, and R. K. Sikes.** 1984. Nosocomial mycobacterial pseudoinfection in a Georgia hospital. Infect. Control **5:**573–576.
9. **Gribetz, A. R., B. Damsker, E. J. Bottone, P. A. Kirshner, and A. S. Teirstein.** 1981. Solitary pulmonary nodules due to nontuberculous mycobacterial infection. Am. J. Med. **70:**39–43.
10. **Guarderas, J., S. Alvarez, and S. L. Berk.** 1986. Progressive pulmonary disease caused by *Mycobacterium gordonae*. South. Med. J. **79:**505–507.
11. **Kubala, E.** 1972. Some aspects of diseases caused by atypical mycobacteria. Scand. J. Respir. Dis. **80(Suppl.):**11–16.
12. **Kumar, U. N., and B. Varkey.** 1980. Pulmonary infection caused by *Mycobacterium gordonae*. Br. J. Dis. Chest **44:**189–192.
13. **Kurnik, P. B., U. Padmanabh, C. Bonatsos, and M. H. Cyna-**

mon. 1983. Case report: *Mycobacterium gordonae* as a human hepato-peritoneal pathogen, with a review of the literature. Am. J. Med. Sci. **285:**45–48.

14. **Lohr, D. C., J. A. Gowken, D. B. Doty, and S. T. Donta.** 1978. *Mycobacterium gordonae* infection of a prosthetic aortic valve. J. Am. Med. Assoc. **293:**1528–1530.

15. **London, R., B. Damsker, E. P. Neibart, B. Knorr, and E. J. Bottone.** 1988. *Mycobacterium gordonae*: an unusual peritoneal pathogen in a patient undergoing continuous ambulatory peritoneal dialysis. Am. J. Med. **85:**703–704.

16. **McIntyre, P., Z. Blacklock, and J. G. McCormack.** 1987. Cutaneous infection with *Mycobacterium gordonae*. J. Infect. **14:**71–78.

17. **Roberts, G. D., N. L. Goodman, L. Heifets, H. W. Larsh, T. H. Lindner, J. K. McClatchy, M. R. McGinnis, S. H. Siddiqi, and P. Wright.** 1983. Evaluation of the BACTEC radiometric method for recovery of mycobacteria and drug susceptibility testing of *Mycobacterium tuberculosis* from acid-fast smear-positive specimens. J. Clin. Microbiol. **18:**689–696.

18. **Shelley, W. B., and A. T. Folkens.** 1984. *Mycobacterium gordonae* infection of the hand. Arch. Dermatol. **120:**1064–1065.

19. **Siddiqi, S. H.** 1989. BACTEC TB System, product and procedure manual. Becton Dickinson Diagnostic Instrument Systems, Towson, Md.

20. **Steere, A. C., J. Corrales, and A. von Graevenitz.** 1979. A Cluster of *Mycobacterium gordonae* isolates from bronchoscopy specimens. Am. Rev. Respir. Dis. **120:**214–216.

21. **Stine, T. M., A. A. Harris, S. Levin, N. Rivera, and R. L. Kaplan.** 1987. A pseudoepidemic due to atypical mycobacteria in a hospital water supply. J. Am. Med. Assoc. **258:**809–811.

22. **Wayne, L. G., and G. P. Kubica.** 1986. The mycobacteria, p. 1447. *In* P. H. A. Sneath, N. S. Mair, M. E. Sharpe, and J. G. Holt (ed.), Bergey's manual of systematic bacteriology, vol. 2. The Williams & Wilkins Co., Baltimore.