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Mycobacterium Growth Indicator Tube Testing in Conjunction with the AccuProbe or the AMPLICOR-PCR Assay for Detecting and Identifying Mycobacteria from Sputum Samples

SATOSHI ICHIYAMA,[1,2]* YOSHITSUGU IINUMA,[3] SADAAKI YAMORI,[3] YOSHINORI HASEGAWA,[2] KAORU SHIMOKATA,[2] AND NOBUO NAKASHIMA[1]

Department of Clinical Laboratory Medicine, Nagoya University Hospital,[1] and The First Department of Internal Medicine,[2] Nagoya University School of Medicine, Showa-ku, Nagoya 466, and Department of Internal Medicine, Chubu National Hospital, Obu 474,[3] Japan

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We have compared the ability of the Mycobacterium Growth Indicator Tube (MGIT) system, a new culture method with an oxygen-sensitive fluorescent sensor, to recover mycobacteria from sputum samples with the abilities of egg-based medium and the Septi-Chek AFB system. We have also assessed the clinical utility of the AccuProbe or the AMPLICOR-PCR assay to directly identify *Mycobacterium tuberculosis* complex and *M. avium-M. intracellulare* complex (MAC) from positive MGITs. From 382 sputum samples, 99 isolates of *M. tuberculosis* complex and 20 isolates of MAC were recovered. The MGIT system had the highest recovery rates for *M. tuberculosis* complex (97.0%) and MAC (100%), compared to recovery rates of 51.5 and 65.0%, respectively, with the egg-based medium and 81.8 and 85.0%, respectively, with the Septi-Chek AFB system. The shortest recovery times were also achieved with the MGIT system: 16.6 days for *M. tuberculosis* complex and 12.0 days for MAC, compared to 27.1 and 20.1 days, respectively, with the egg-based medium and 21.4 and 13.2 days, respectively, with the Septi-Chek AFB system. The AccuProbe identified 74 (77.1%) of the 96 *M. tuberculosis* complex-positive MGITs and 17 (85.0%) of the 20 MAC-positive vials. The AMPLICOR system correctly identified 94 (97.9%) of the 96 *M. tuberculosis* complex-positive MGITs and all 20 MAC-positive vials. Therefore, the MGIT system used in conjunction with the AMPLICOR system is a rapid and sensitive method for detecting and identifying *M. tuberculosis* complex and MAC isolates from sputum samples.

The diagnosis of tuberculosis continues to depend on microscopy and culture. Microscopic examination by either Ziehl-Neelsen or auramine-rhodamine staining is insensitive. Culture on egg-based Lowenstein-Jensen or agar-based Middlebrook 7H11 medium is time-consuming (requiring 3 to 8 weeks), and the sensitivities of solid media are unsatisfactory. More sensitive liquid media, such as those included with the radiometric BACTEC 460TB system (Becton Dickinson Microbiology Systems, Sparks, Md.) (2, 20) and the nonradiometric Septi-Chek AFB system (Becton Dickinson) (2, 16, 18), are used in most clinical laboratories to recover mycobacteria. Species-specific DNA probes, such as the radiometric Gen-Probe and nonradiometric AccuProbe (Gen-Probe Inc., San Diego, Calif.), have dramatically shortened the time required to identify *Mycobacterium tuberculosis* complex, *M. avium*, *M. intracellulare*, *M. kansasii*, and *M. gordonae* from colonies of mycobacteria (15, 25, 31).

Amplification techniques such as PCR provide rapid methods for directly detecting mycobacterial DNA or RNA from clinical samples (1, 4–6, 8, 10–12, 17, 19, 22–24, 26, 27, 29, 32–35, 37). Recently, two commercially available kits, the rRNA amplification-based Gen-Probe Amplified Mycobacterium Tuberculosis Direct Test system (Gen-Probe) (1, 17, 19, 22, 24, 34, 35) and the PCR-based Roche AMPLICOR MYCOBACTERIUM system (Roche, Basel, Switzerland) (4–6, 8, 11, 12, 17, 23, 32, 33, 35, 37), have provided the speed, reliability, and requisite diagnostic capability for the direct detection and identification of mycobacteria.

A new liquid medium system used for culture and susceptibility testing, the Mycobacterium Growth Indicator Tube (MGIT; Becton Dickinson) (3, 28, 36) system, has recently been developed. This system can detect the growth of mycobacteria more quickly than conventional solid media by using an oxygen-sensitive fluorescent sensor. Few studies have evaluated the clinical use of this system and compared it with other commercially available liquid culture systems. The purpose of this study was to compare the sensitivity and rapidity of the MGIT system with those of conventional solid and liquid media for the detection of mycobacteria from clinical samples. We also assessed the usefulness of the AccuProbe and the AMPLICOR system for the direct identification of mycobacteria from positive MGITs. Finally, we propose a rapid and sensitive method for detecting and identifying mycobacteria in clinical laboratories.

MATERIALS AND METHODS

Sample processing. Clinical samples were obtained from patients suspected of having mycobacterial infection of the lung and from those undergoing antituberculosis chemotherapy at Chubu National Hospital, Obu, Aichi Prefecture, Japan, from June 1995 through March 1996. A total of 382 sputum samples from 141 patients were examined in this study. All samples were decontaminated by treating them with an equal volume of *N*-acetyl-L-cysteine–NaOH (final concentration, 2%) for 15 min at room temperature and were then neutralized with sterile 0.067 M phosphate buffer (pH 6.8). After centrifugation at 3,000 × *g* for 15 min, the sediment was resuspended in 2.0 ml of phosphate buffer. All specimens were stained with auramine-rhodamine stain to detect acid-fast bacilli (AFB). Each 0.5 ml of the decontaminated suspension was inoculated into both an MGIT system and a Septi-Chek AFB system. An additional 0.1 ml of the suspension was inoculated onto an egg-based Ogawa medium at 37°C. Mycobacterial growth was evaluated daily (except for Saturday and Sunday) for 8 weeks. Ogawa medium is commonly used as an alternative to Lowenstein-Jensen me-

* Corresponding author. Mailing address: Department of Clinical Laboratory Medicine, Nagoya University Hospital, 65 Tsurumai-cho, Showa-ku, Nagoya 466, Japan. Phone: (81)-52-744-2614. Fax: (81)-52-744-2613.

TABLE 1. Recovery of mycobacteria with egg-based Ogawa medium, Septi-Chek AFB system, and MGIT system

Mycobacterium (no. of isolates)	No. (%) of isolates recovered		
	Egg medium	Septi-Chek AFB system	MGIT system
MTC[a] (99)	51 (51.5)	81 (81.8)	96 (97.0)
MAC (20)	13 (65.0)	17 (85.0)	20 (100)
Mycobacterium spp.[b] (4)	1 (25.0)	3 (75.0)	2 (50.0)
Total (123)	65 (52.8)	101 (82.1)	118 (95.9)

[a] MTC, *M. tuberculosis* complex.
[b] *Mycobacterium* spp., mycobacteria other than *M. tuberculosis* complex or MAC.

dium in Japan. Cultures were determined to be positive when the presence of AFB was confirmed by Ziehl-Neelsen staining. The mycobacterial colonies present on a slant of Ogawa medium or a Middlebrook 7H11 agar paddle of the Septi-Chek AFB system were identified with the AccuProbe. When only the MGIT system was positive, the organism was identified after it formed colonies on an Ogawa medium. All of the positive MGITs were tested with the AccuProbe and the AMPLICOR system to determine the abilities of the AccuProbe and the AMPLICOR system to directly identify the organisms in the MGITs.

MGIT system. The MGIT system contains 4.0 ml of an enriched Middlebrook 7H9 broth with 0.25% glycerol. A fluorescent indicator quenched by oxygen is embedded in silicone on the bottom of a round-bottom tube (16 by 100 mm). When the growing mycobacteria are consuming oxygen, the fluorescence can easily be detected visually with the aid of 365-nm UV light. A total of 0.5 ml of the decontaminated suspension was inoculated into an MGIT to which 0.5 ml of MGIT oleic acid-albumin-dextrose-catalase enrichment and 0.1 ml of MGIT polymyxin B, azlocillin, nalidixic acid, trimethoprim, and amphotericin B antibiotics had previously been added. Each MGIT was incubated at 37°C and was read with a UV light daily (except for Saturday and Sunday) for 8 weeks. Positive vials were immediately frozen on the day that the vial became positive and were stored at −80°C until identification tests were performed by DNA-based methods.

Identification of organisms from positive MGITs by DNA-based methods. After the presence of AFB was confirmed by Ziehl-Neelsen staining, the mycobacteria in the positive MGITs which had been stored at −80°C were directly identified from the broth with the AccuProbe and the AMPLICOR system. Two 1.0-ml aliquots of broth from the AFB-positive MGITs were centrifuged at 3,000 × *g* for 15 min. The supernatants were decanted, and the pellets were used in the two identification tests according to the manufacturers' instructions. The species-specific probes that were used were directed toward *M. tuberculosis* complex and *M. avium-M. intracellulare* complex (MAC) for the AccuProbe and toward *M. tuberculosis* complex, *M. avium*, and *M. intracellulare* with the AMPLICOR system. The AMPLICOR system with probes toward *M. tuberculosis* complex, *M. avium*, and *M. intracellulare* is commercially available in Japan.

Statistical analysis. Recovery rates and the number of days required to recover mycobacteria with each of the three culture systems were evaluated by the McNemar modification of the chi-square test and the paired Wilcoxon test, respectively. The rates of organism identification from positive MGITs with the AccuProbe and the AMPLICOR system were also evaluated by the McNemar modification of the chi-square test.

RESULTS

Of the 382 sputum samples from the 141 patients, 71 (18.6%) were smear positive and 123 (32.2%) were culture positive with Ogawa egg medium, the Septi-Chek AFB system, and/or the MGIT system. Of the 123 isolates identified, 99 (80.5%) were *M. tuberculosis* complex, 20 (16.3%) were MAC, and 4 (3.3%) were mycobacteria other than *M. tuberculosis* complex or MAC. Of the 71 smear-positive samples, 7 were culture negative by all three culture methods. Six, two, and three samples tested with Ogawa medium, the Septi-Chek AFB system, and the MGIT system, respectively, were contaminated.

Table 1 presents the abilities of the three culture methods to recover *M. tuberculosis* complex, MAC, and other mycobacteria. Recovery rates were calculated by dividing the number of isolates recovered by each method by the total number of isolates recovered by any of the three methods. The MGIT system had the highest recovery rates for both *M. tuberculosis* complex (97.0%) and MAC (100%). The recovery rates of Ogawa medium and the Septi-Chek AFB system were 51.5 and 81.8%, respectively, for *M. tuberculosis* complex and 65.0 and 85.0%, respectively, for MAC. There were significant differences between the MGIT system and Ogawa medium in terms of the recovery rates for both *M. tuberculosis* complex ($P < 0.0001$) and MAC ($P = 0.0233$). The difference between the two liquid culture systems in the recovery rates was statistically significant for *M. tuberculosis* complex ($P = 0.0023$) but was insignificant ($P = 0.2482$) for MAC. The overall recovery rates of all mycobacteria isolated were 52.8, 82.1, and 95.9% for Ogawa egg medium, the Septi-Chek AFB system, and the MGIT system, respectively.

TABLE 2. Average number of days required to recover mycobacteria with egg-based Ogawa medium, Septi-Chek AFB system, and MGIT system

Mycobacterium	No. of days required for recovery (no. of isolates)		
	Egg medium	Septi-Chek AFB system	MGIT system
MTC[a]	27.1 (51)	21.4 (81)	16.6 (96)
MAC	20.1 (13)	13.2 (17)	12.0 (20)

[a] MTC, *M. tuberculosis* complex.

Table 2 indicates the average number of days required to recover mycobacteria by each of the three methods. The shortest detection times were achieved with the MGIT system for both *M. tuberculosis* complex and MAC. For *M. tuberculosis* complex, the MGIT system required 16.6 days, whereas Ogawa medium required 27.1 days ($P < 0.0001$) and the Septi-Chek AFB system required 21.4 days ($P < 0.0001$). For MAC, 12.0 days was required for recovery by the MGIT system, whereas 20.1 days was required by Ogawa medium ($P = 0.0014$) and 13.2 days was required by the Septi-Chek AFB system ($P = 0.0690$). These statistical analyses were done with samples positive by both of the culture methods evaluated. Table 3 presents the average number of days for recovery according to the smear results for the samples. The MGIT system recovered *M. tuberculosis* complex from the smear-positive samples in 11.5 days and MAC from the smear-positive samples in 7.8 days. For the smear-negative samples, the MGIT system required 22.5 days to recover *M. tuberculosis* complex and 16.2 days to recover MAC.

Table 4 presents the sensitivities of the AccuProbe and the AMPLICOR system for direct identification from the positive MGITs according to the smear results for the samples. The AccuProbe identified *M. tuberculosis* complex in 43 (84.3%) of the 51 smear-positive samples and 31 (68.9%) of the 45 smear-negative samples and identified MAC in 10 (100%) of the 10 smear-positive samples and 7 (70.0%) of the 10 smear-negative samples. On the other hand, the AMPLICOR system correctly

TABLE 3. Average number of days required to recover mycobacteria with egg-based Ogawa medium, Septi-Chek AFB system, and MGIT system according to smear results

Smear result/ mycobacterium	No. of days required for recovery (no. of isolates)		
	Egg medium	Septi-Chek AFB system	MGIT system
Positive/MTC[a]	22.0 (37)	17.0 (48)	11.5 (51)
Negative/MTC	40.6 (14)	27.9 (33)	22.5 (45)
Positive/MAC	15.6 (8)	11.8 (10)	7.8 (10)
Negative/MAC	27.2 (5)	15.3 (7)	16.2 (10)

[a] MTC, *M. tuberculosis* complex.

TABLE 4. Sensitivity of the AccuProbe and the AMPLICOR system for directly identifying mycobacteria from positive MGITs

Smear result/mycobacterium (no. of isolates)	No. (%) of isolates correctly identified	
	AccuProbe	AMPLICOR system
Positive/MTC[a] (51)	43 (84.3)	51 (100)
Negative/MTC (45)	31 (68.9)	43 (95.6)
Positive/MAC (10)	10 (100)	10 (100)
Negative/MAC (10)	7 (70.0)	10 (100)
Total (116)	91 (78.4)	114 (98.3)

[a] MTC, *M. tuberculosis* complex.

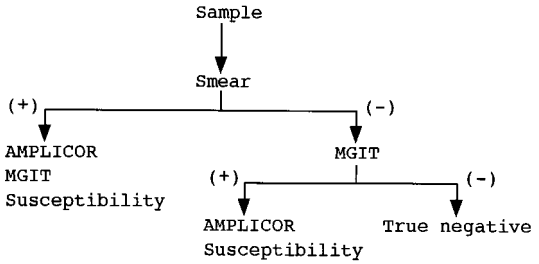

FIG. 1. Proposed combination test protocol for detecting and identifying mycobacteria from sputum samples. +, positive result; −, negative result.

identified *M. tuberculosis* complex isolates in 94 (97.9%) of the 96 *M. tuberculosis* complex-positive MGITs and MAC in all 20 MAC-positive MGITs. The organisms in two MGITs false negative for *M. tuberculosis* complex were also unidentified with the AccuProbe. The difference between the two tests in the overall rate of identification of *M. tuberculosis* complex and MAC was statistically significant ($P < 0.0001$).

DISCUSSION

The resurgence of mycobacterial infections in association with human immunodeficiency virus infection has prompted the development of rapid methods for the detection and identification of the causative mycobacteria. The clinical utility of the AMPLICOR system has been evaluated by many investigators (4–6, 8, 11, 12, 17, 23, 32, 33, 35, 37). For smear-positive specimens, the sensitivity and specificity of this system were excellent for the detection of *M. tuberculosis* complex isolates. On the other hand, several reports have indicated that this system has unsatisfactory sensitivities for smear-negative specimens, ranging from approximately 40 to 70% (5, 6, 11, 12, 23, 37). Because of the low sensitivity for smear-negative specimens and the high cost of this system, further studies are needed to determine the most clinically relevant use of this system for smear-negative specimens. Culture methods are still required because of potential false-negative results of the PCR assay and the need to determine the susceptibilities of the isolates.

This study showed the higher sensitivities and shorter detection times of the Septi-Chek system and the MGIT system compared to those of Ogawa medium. The study design could account for the decreased sensitivity of and rapidity of detection with Ogawa medium, since this medium received 0.1 ml of decontaminated specimen whereas each of the two liquid media was inoculated with 0.5-ml aliquots. Use of the MGIT system, a novel system that uses liquid culture medium, is simple and flexible, and the system requires no special equipment or radioactive reagents. An incubated vial can be screened at any time for the presence of fluorescence, and AFB can be detected before the liquid medium becomes turbid. In this study, the MGIT system was found to be superior to the Septi-Chek AFB system with regard to the rate of recovery and the time to detection of *M. tuberculosis* complex and MAC from sputum samples. Since the Septi-Chek AFB bottle is screened by examining the turbidity of the liquid medium, positive bottles may be overlooked if the turbidity is faint. The Septi-Chek system has already demonstrated great recovery rates equivalent to those of the BACTEC 460TB system, but it failed to show detection times equivalent to those of the BACTEC 460TB system (2, 18). The higher sensitivity of and shorter time to detection with the MGIT system were considered to result from the use of an oxygen-sensitive fluorescent compound. However, the MGIT system does not include a plate which generates mycobacterial colonies to be identified with DNA probes.

Several investigators have used the Gen-Probe or the AccuProbe to directly identify mycobacteria from positive BACTEC 460TB vials (7, 9, 13, 14, 21, 30). Rates of identification of *M. tuberculosis* complex, MAC, *M. kansasii*, and *M. gordonae* from positive vials with the probes have varied by species or by the number of mycobacterial organisms in the vials, showing sensitivities of 47 to 100% for *M. tuberculosis* complex and 78.5 to 100% for MAC. Another report has demonstrated the usefulness of the AMPLICOR system for directly identifying mycobacteria from positive BACTEC 460TB vials, showing a sensitivity and a specificity of 98 and 100%, respectively (37). Although combination tests have been shown to be useful, the BACTEC 460TB system is not acceptable for use by all clinical laboratories because of the problems with radioactive waste. A recent report has indicated the excellent sensitivity of the MGIT system, equivalent to that of the BACTEC 460TB system, for detecting mycobacteria from clinical specimens (3). Few data are available concerning the use of the MGIT system in conjunction with DNA probes or amplification systems for the rapid detection and identification of mycobacteria.

Given these concerns, we have designed the most appropriate application of the DNA-based assays (probe and amplification assays) and culture systems for the detection and identification of mycobacteria with regard to sensitivity, rapidity, and the use of nonradioactive substances. Our study showed that the MGIT system has higher recovery rates and shorter detection times than the egg-based medium and the Septi-Chek AFB system for both *M. tuberculosis* complex and MAC. The sensitivities of the AMPLICOR system in directly identifying *M. tuberculosis* complex and MAC from the positive MGITs were excellent. Figure 1 illustrates a proposed combination test protocol. For the smear-positive samples, the AMPLICOR system is used for the rapid diagnosis of *M. tuberculosis* complex and MAC. The sample is simultaneously inoculated into the MGIT system for the identification of mycobacteria other than *M. tuberculosis* complex or MAC and for susceptibility testing of the isolate. When the smear result is negative, the sample is initially inoculated into the MGIT system. If the culture result with the MGIT system is positive, the AMPLICOR system is adopted to directly identify *M. tuberculosis* complex or MAC from the positive MGIT. The average numbers of days required to detect and identify mycobacteria are estimated to be 22.5 days for *M. tuberculosis* complex and 16.2 days for MAC. Since the infectivity of patients with tuberculosis whose sputum samples are smear negative is low, the incubation times for the MGIT system required to provide a diagnosis for these patients are considered to be acceptable. Our combination protocol may provide a useful method for screening and the subsequent identification testing of smear-

negative samples, because the U.S. Food and Drug Administration does not recommend the use of the amplification tests to directly screen smear-negative samples at this time.

In summary, the MGIT system, when used in conjunction with the AMPLICOR system, is a rapid and sensitive method that does not use radioactive compounds and that is useful for the detection and identification of *M. tuberculosis* complex and MAC. Therefore, we recommend the use of the combined protocol for routine testing for mycobacterial infections in clinical laboratories.

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REFERENCES

1. **Abe, C., K. Hirano, M. Wada, Y. Kazumi, M. Takahashi, Y. Fukasawa, T. Yoshimura, C. Miyagi, and S. Goto.** 1993. Detection of *Mycobacterium tuberculosis* in clinical specimens by polymerase chain reaction and Gen-Probe Amplified Mycobacterium Tuberculosis Direct Test. J. Clin. Microbiol. **31:** 3270–3274.
2. **Abe, C., S. Hosojima, Y. Fukasawa, Y. Kazumi, M. Takahashi, K. Hirano, and T. Mori.** 1992. Comparison of MB-Check, BACTEC, and egg-based media for recovery of mycobacteria. J. Clin. Microbiol. **30:**878–881.
3. **Badak, F. Z., D. L. Kiska, S. Setterquist, C. Hartley, M. A. O'Connell, and R. L. Hopfer.** 1996. Comparison of Mycobacteria Growth Indicator Tube with BACTEC 460 for detection and recovery of mycobacteria from clinical specimens. J. Clin. Microbiol. **34:**2236–2239.
4. **Beavis, K. G., M. B. Lichty, D. L. Jungkind, and O. Giger.** 1995. Evaluation of Amplicor PCR for direct detection of *Mycobacterium tuberculosis* from sputum specimens. J. Clin. Microbiol. **33:**2582–2586.
5. **Bennedsen, J., V. O. Thomsen, G. E. Pfyffer, G. Funke, K. Feldmann, A. Beneke, P. A. Jenkins, M. Hegginbothom, A. Fahr, M. Hengstler, G. Cleator, P. Klapper, and E. G. L. Wilkins.** 1996. Utility of PCR in diagnosing pulmonary tuberculosis. J. Clin. Microbiol. **34:**1407–1411.
6. **Bergmann, J. S., and G. L. Woods.** 1996. Clinical evaluation of the Roche AMPLICOR PCR *Mycobacterium tuberculosis* test for detection of *M. tuberculosis* in respiratory specimens. J. Clin. Microbiol. **34:**1083–1085.
7. **Body, B. A., N. G. Warren, A. Spicer, D. Henderson, and M. Chery.** 1990. Use of Gen-Probe and Bactec for rapid isolation and identification of mycobacteria. Correlation of probe results with growth index. Am. J. Clin. Pathol. **93:**415–420.
8. **Carpentier, E., B. Drouillard, M. Dailloux, D. Moinard, E. Vallee, B. Dutilh, J. Maugein, E. Bergogne-Berezin, and B. Carbonnelle.** 1995. Diagnosis of tuberculosis by Amplicor *Mycobacterium tuberculosis* test: a multicenter study. J. Clin. Microbiol. **33:**3106–3110.
9. **Chapin-Robertson, K., S. Dahlberg, S. Waycott, J. Corrales, C. Kontnic, and S. C. Edberg.** 1993. Detection and identification of mycobacterium directly from BACTEC bottles by using a DNA-rRNA probe. Diagn. Microbiol. Infect. Dis. **17:**203–207.
10. **Clarridge, J. E., III, R. M. Shawar, T. M. Shinnick, and B. B. Plikaytis.** 1993. Large-scale use of polymerase chain reaction for detection of *Mycobacterium tuberculosis* in a routine mycobacteriology laboratory. J. Clin. Microbiol. **31:**2049–2056.
11. **D'Amato, R. F., A. A. Wallman, L. H. Hochstein, P. M. Colaninno, M. Scardamaglia, E. Ardila, M. Ghouri, K. Kim, R. C. Patel, and A. Miller.** 1995. Rapid diagnosis of pulmonary tuberculosis by using Roche AMPLICOR *Mycobacterium tuberculosis* PCR test. J. Clin. Microbiol. **33:**1832–1834.
12. **Devallois, A., E. Legrand, and N. Rastogi.** 1996. Evaluation of Amplicor MTB test as adjunct to smears and culture for direct detection of *Mycobacterium tuberculosis* in the French Caribbean. J. Clin. Microbiol. **34:**1065–1068.
13. **Ellner, P. D., T. E. Kiehn, R. Cammarata, and M. Hosmer.** 1988. Rapid detection and identification of pathogenic mycobacteria by combining radiometric and nucleic acid probe methods. J. Clin. Microbiol. **26:**1349–1352.
14. **Evans, K. D., A. S. Nakasone, P. A. Sutherland, L. A. de la Maza, and E. M. Perterson.** 1992. Identification of *Mycobacterium tuberculosis* and *Mycobacterium avium-M. intracellulare* directly from primary BACTEC cultures by using acridinium-ester-labeled DNA probes. J. Clin. Microbiol. **30:**2427–2431.
15. **Goto, M., S. Oka, K. Okuzumi, S. Kimura, and K. Shimada.** 1991. Evaluation of acridinium-ester-labeled DNA probes for identification of *Mycobacterium tuberculosis* and *Mycobacterium avium-Mycobacterium intracellulare* complex in culture. J. Clin. Microbiol. **29:**2473–2476.
16. **Ichiyama, S., K. Shimokata, J. Takeuchi, and the Aichi Mycobacteriosis Research Group.** 1993. Comparative study of a biphasic culture system (Roche MB Check system) with a conventional egg medium for recovery of mycobacteria. Tubercle Lung Dis. **74:**338–341.
17. **Ichiyama, S., Y. Iinuma, Y. Tawada, S. Yamori, Y. Hasegawa, K. Shimokata, and N. Nakashima.** 1996. Evaluation of Gen-Probe Amplified Mycobacterium Tuberculosis Direct Test and Roche PCR-microwell plate hybridization method (AMPLICOR MYCOBACTERIUM) for direct detection of mycobacteria. J. Clin. Microbiol. **34:**130–133.
18. **Isenberg, H. D., R. F. D'Amato, L. Heifets, P. R. Murray, M. Scardamaglia, M. C. Jacobs, P. Alperstein, and A. Niles.** 1991. Collaborative feasibility study of a biphasic system (Roche Septi-Chek AFB) for rapid detection and isolation of mycobacteria. J. Clin. Microbiol. **29:**1719–1722.
19. **Jonas, V., M. J. Alden, J. I. Curry, K. Kamisango, C. A. Knott, R. Lankford, J. M. Wolfe, and D. F. Moore.** 1993. Detection and identification of *Mycobacterium tuberculosis* directly from sputum sediments by amplification of rRNA. J. Clin. Microbiol. **31:**2410–2416.
20. **Kirihara, J. M., S. L. Hillier, and M. B. Coyle.** 1985. Improved detection times for *Mycobacterium avium* complex and *Mycobacterium tuberculosis* with the BACTEC radiometric system. J. Clin. Microbiol. **22:**841–845.
21. **Metchock, B., and L. Diem.** 1995. Algorithm for use of nucleic acid probes for identifying *Mycobacterium tuberculosis* from BACTEC 12B bottles. J. Clin. Microbiol. **33:**1934–1937.
22. **Miller, N., S. G. Hernandez, and T. J. Cleary.** 1994. Evaluation of Gen-Probe Amplified Mycobacterium Tuberculosis Direct Test and PCR for direct detection of *Mycobacterium tuberculosis* in clinical specimens. J. Clin. Microbiol. **32:**393–397.
23. **Moore, D. F., and J. I. Curry.** 1995. Detection and identification of *Mycobacterium tuberculosis* directly from sputum sediments by Amplicor PCR. J. Clin. Microbiol. **33:**2686–2691.
24. **Moore, D. F., J. I. Curry, C. A. Knott, and V. Jonas.** 1996. Amplification of rRNA for assessment of treatment response of pulmonary tuberculosis patients during antimicrobial therapy. J. Clin. Microbiol. **34:**1745–1749.
25. **Musial, C. E., L. S. Tice, L. Stockman, and G. D. Roberts.** 1988. Identification of mycobacteria from culture by using the Gen-Probe rapid diagnostic system for *Mycobacterium avium* complex and *Mycobacterium tuberculosis* complex. J. Clin. Microbiol. **26:**2120–2123.
26. **Nolte, F. S., B. Metchock, J. E. McGowan, Jr., A. Edwards, O. Okwumabua, C. Thurmond, P. S. Mitchell, B. Plikaytis, and T. Shinnick.** 1993. Direct detection of *Mycobacterium tuberculosis* in sputum by polymerase chain reaction and DNA hybridization. J. Clin. Microbiol. **31:**1777–1782.
27. **Noordhoek, G. T., A. H. J. Kolk, G. Bjune, D. Catty, J. W. Dale, P. E. M. Fine, P. Godfrey-Faussett, S. N. Cho, T. Shinnick, S. B. Svenson, S. Wilson, and J. D. A. van Embden.** 1994. Sensitivity and specificity of PCR for detection of *Mycobacterium tuberculosis*: a blind comparison study among seven laboratories. J. Clin. Microbiol. **32:**277–284.
28. **Palaci, M., S. Y. M. Ueki, D. N. Sato, M. A. S. Telles, M. Curcio, and E. A. M. Silva.** 1996. Evaluation of Mycobacteria Growth Indicator Tube for recovery and drug susceptibility testing of *Mycobacterium tuberculosis* isolates from respiratory specimens. J. Clin. Microbiol. **34:**762–764.
29. **Pfyffer, G. E., P. Kissling, R. Wirth, and R. Weber.** 1994. Direct detection of *Mycobacterium tuberculosis* complex in respiratory specimens by a target-amplified test system. J. Clin. Microbiol. **32:**918–923.
30. **Reisner, B. S., A. M. Gatson, and G. L. Woods.** 1994. Use of Gen-Probe AccuProbes to identify *Mycobacterium avium* complex, *Mycobacterium tuberculosis* complex, *Mycobacterium kansasii*, and *Mycobacterium gordonae* directly from BACTEC TB broth cultures. J. Clin. Microbiol. **32:**2995–2998.
31. **Saito, H., H. Tomioka, K. Sato, H. Tasaka, and D. J. Dawson.** 1990. Identification of various serovar strains of *Mycobacterium avium* complex by using DNA probes specific for *Mycobacterium avium* and *Mycobacterium intracellulare*. J. Clin. Microbiol. **28:**1694–1697.
32. **Schirm, J., L. A. B. Oostendorp, and J. G. Mulder.** 1995. Comparison of Amplicor, in-house PCR, and conventional culture for detection of *Mycobacterium tuberculosis* in clinical samples. J. Clin. Microbiol. **33:**3221–3224.
33. **Sergmann, J. S., and G. L. Woods.** 1996. Clinical evaluation of the Roche AMPLICOR PCR *Mycobacterium tuberculosis* test for detection of *M. tuberculosis* in respiratory specimens. J. Clin. Microbiol. **34:**1083–1085.
34. **Vlaspolder, F., P. Singer, and C. Roggeveen.** 1995. Diagnostic value of an amplification method (Gen-Probe) compared with that of culture for diagnosis of tuberculosis. J. Clin. Microbiol. **33:**2699–2703.
35. **Vuorinen, P., A. Miettinen, R. Vuento, and O. Hällström.** 1995. Direct detection of *Mycobacterium tuberculosis* complex in respiratory specimens by Gen-Probe Amplified Mycobacterium Tuberculosis Direct Test and Roche Amplicor Mycobacterium Tuberculosis Test. J. Clin. Microbiol. **33:**1856–1859.
36. **Walters, S. B., and B. A. Hanna.** 1996. Testing of susceptibility of *Mycobacterium tuberculosis* to isoniazid and rifampin by Mycobacterium Growth Indicator Tube method. J. Clin. Microbiol. **34:**1565–1567.
37. **Wobeser, W. L., M. Krajden, J. Conly, H. Simpson, B. Yim, M. D'Costa, M. Fuksa, C. Hian-Cheong, M. Patterson, A. Phillips, R. Bannatyne, A. Haddad, J. L. Brunton, and S. Krajden.** 1996. Evaluation of Roche Amplicor PCR assay for *Mycobacterium tuberculosis*. J. Clin. Microbiol. **34:**134–139.