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Multicenter Evaluation of the MB/BACT System for Susceptibility Testing of *Mycobacterium tuberculosis*

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The reliability of the MB/BACT system for susceptibility testing of *Mycobacterium tuberculosis* to pyrazinamide, rifampin, isoniazid, streptomycin, and ethambutol was compared to the BACTEC 460TB system. The proportion method was used to resolve discrepant results by an independent arbiter. Two interpretative methods were used, with an undiluted control (direct control) and a diluted control (10^{-1} control). As no significant difference was observed between the two controls, the method with the direct control was adopted as the most accurate one. One hundred sixty-six strains were tested, with an overall agreement of 98.3%. After resolution of the 18 discrepant results by the proportion method, the sensitivity and specificity of the MB/BACT system were 100% for rifampin, isoniazid, and pyrazinamide. For ethambutol, sensitivity was 92.3% at the critical concentration and 33% at the high concentration, and specificity was 100% at both concentrations. For streptomycin, sensitivity was 100% at the critical concentration and 80% at the high concentration, and specificity was 98.6% at the critical concentration and 100% at the high concentration. The rifampin, isoniazid, streptomycin, and ethambutol susceptibility test results were obtained in 6.6 days with the MB/BACT versus 5 days with the BACTEC 460TB. The pyrazinamide susceptibility test results were obtained in 7.8 days with the MB/BACT, versus 6.7 days with the BACTEC 460TB. These data demonstrate that the fully automated MB/BACT system is a very reliable method for rapid susceptibility testing of *M. tuberculosis* against rifampin, isoniazid, and pyrazinamide. Sensitivity results have to be improved for ethambutol and streptomycin, especially at the high concentration.

With three million deaths and ten million people infected each year, tuberculosis is still the infectious disease with the highest morbidity and mortality. In addition, multidrug-resistant *Mycobacterium tuberculosis* strains have been emerging worldwide in both high- and low-income countries. The need for rapid methods of diagnosis and determination of drug susceptibility is particularly important. The Centers for Disease Control and Prevention in Atlanta, Ga., recommend that susceptibility test results for *M. tuberculosis* complex isolates be available 28 to 30 days from receipt of a specimen in the laboratory (16). The most widely used methods for antimycobacterial susceptibility testing are the proportion method on solid medium and the radiometric procedure on liquid broth. The former procedure cannot provide results before 21 days of inoculation. The radiometric BACTEC 460TB requires less than 10 days of incubation before results are available (13). The BACTEC 460TB system, however, is semiautomated and entails disposal of a radioactive substance.

Recently, new liquid medium-based systems have been evaluated for nonradiometric susceptibility testing of *M. tuberculosis*, such as the ESP Culture System II (AccuMed International, Westlake, Ohio), MB Redox (Biotest, Dreieich, Germany), the Mycobacteria Growth Indicator Tube (manual MGIT or fully automated BACTEC MGIT 960 system; Becton Dickinson Microbiology System, Sparks, Md.), and the MB/BACT (BioMerieux). Previous evaluations of these new systems report good overall agreement of antimycobacterial susceptibility testing results with those obtained by established methods (2–6, 11, 14, 17–18, 20). Until recently, the radiometric procedure was the only rapid method available to test pyrazinamide susceptibility among isolates of *Mycobacterium tuberculosis*. Three recent studies reported the results of susceptibility testing of *M. tuberculosis* to pyrazinamide performed on the ESP system and on the BACTEC MGIT 960 system (1, 8, 12).

In this multicenter study, we have evaluated the reproducibility and reliability of the MB/BACT system for testing of *M. tuberculosis* susceptibility to pyrazinamide, rifampin, isoniazid, streptomycin, and ethambutol, known as the PRISE drugs. Most previous studies evaluating the MB/BACT system tested the MB/BACT kit previously evaluated in Spanish and Italian studies with one concentration per drug: 1 μg/ml for isoniazid, rifampin, and streptomycin, and 2 μg/ml for ethambutol (6, 17, 20). The susceptibility of *M. tuberculosis* to pyrazinamide was evaluated in one study by the MB/BACT system with an additional procedure not included in the kit (final concentration of 50 μg/ml) (17). Our study is the first evaluation of the American MB/BACT kit, which contains lower critical concentrations for isoniazid, rifampin, and streptomycin (0.09 μg/ml, 0.9 μg/ml, and 0.45 μg/ml, respectively) and higher critical concentrations for ethambutol (3.5 μg/ml) and for pyrazin-

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amide (200 µg/ml). Higher concentrations (isoniazid, 0.4 µg/ml; ethambutol, 7.0 µg/ml; and streptomycin, 0.9 µg/ml) were used for resistant strains at the critical concentration to evaluate the level of resistance and to compare its detection with the radiometric procedure and the proportion method.

The novel mycobacterial susceptibility kit contains standardized acidified pyrazinamide vials for pyrazinamide testing. Most of the studies evaluating the MB/BACT system used one drug-free control vial diluted 1:100, as with the BACTEC 460TB procedure. The procedure used in this study was a variation of the principles employed by the proportion on solid medium and broth radiometric methods. Two interpretative methods were used, one with a 10^{-1} control and one with an undiluted control (named the direct control), and the most accurate was adopted. The results were compared to those obtained by the radiometric procedure. An additional site acting as an independent arbiter resolved discrepant results by testing the strains with the proportion method.

MATERIALS AND METHODS

Evaluation sites. Susceptibility testing results were generated by four centers, the Department of Medical Microbiology, University of Nantes, Nantes, France (center 1), the Department of Medical Microbiology, Bel-Air Hospital, Thionville, France (center 2), the Department of Clinical Microbiology, Institute for Infectious Diseases, University of Berne, Berne, Switzerland (center 3), and the Department of Medical Microbiology, Cantonal Hospital, Lucerne, Switzerland (center 4). A fifth laboratory, the National Reference Center for Mycobacteria, Pasteur Institute, Paris, France (center 5), acted as an arbiter for the resolution of discrepant results.

Strains. A total of 166 *M. tuberculosis* strains were evaluated in this study. The strains were fresh clinical isolates grown in MB/BACT or selected from the culture collections. These strains were grown on Löwenstein-Jensen (LJ) medium prior to inoculation to the MB/BACT medium. Accuprobe culture confirmation kits (GenProbe, San Diego, Calif.) and biochemical methods were used for identification.

Preparation of inocula. Mycobacterial susceptibility testing with PRISE drugs was performed directly from the positive MB bottle for fresh clinical isolates and through an intermediate MB seed bottle for strains initially grown on LJ medium. Colonies were suspended in Middlebrook 7H9 broth (adjusted to a McFarland standard of 0.5). This suspension was used to inoculate the MB/BACT seed bottle.

(i) RISE panel. A positive specimen bottle was used as the inoculum suspension for susceptibility testing without adjustment if the bottle was continually incubated for no more than 60 h after the positive signal or if pulled within 60 h of the positive signal and refrigerated for no more than 5 days.

(ii) Pyrazinamide panel. A positive specimen bottle was used as the inoculum suspension for susceptibility testing without adjustment if the bottle was continually incubated for no more than 36 h after the positive signal. Cultures from bottles that were continuously incubated from 36 to 60 h after the positive signal were diluted 1:2 in sterile 7H9 broth to prepare the inoculum suspension. Bottles which had been positive for more than 60 h first had to be subcultured again into new MB/BACT medium.

Growth controls and drug-containing bottles (see below) were inoculated with 0.5 ml.

MB/BACT growth control. Two different growth controls were used: a direct growth control, inoculated with the same number of organisms as the drug-containing bottles, and a diluted control, inoculated with 10-fold fewer organisms than the drug-containing bottles. The direct growth control and 10^{-1} RISE control bottles were prepared by adding 0.5 ml of restoring fluid and 0.5 ml of inoculum suspension or 0.5 ml of the inoculum suspension diluted 1:10, respectively. The direct growth control and 10^{-1} pyrazinamide control bottles were prepared by adding 0.5 ml of reconstitution fluid plus 2 ml of acidifying reagent, and 0.5 ml of inoculum suspension or 0.5 ml of the inoculum suspension diluted 1:10, respectively.

Drug solutions. Each of the lyophilized antibiotics was reconstituted with 6.0 ml of restoring fluid (RISE) or with 6.0 ml of reconstitution fluid (pyrazinamide). Part of the reconstituted antibiotic (drug stock solution) was added to an MB/BACT bottle. The pyrazinamide antibiotic bottle was inoculated with 2 ml of

acidifying reagent. Two concentrations were used for isoniazid, streptomycin, and ethambutol, a low concentration (named the critical concentration) and a high concentration, as recommended previously (7). The final critical and high concentrations were 0.09 and 0.4 µg/ml, respectively, for isoniazid, 0.45 and 0.9 µg/ml, respectively, for streptomycin, and 3.5 and 7.0 µg/ml, respectively, for ethambutol. Rifampin and pyrazinamide were tested at critical concentrations of 0.9 µg/ml and 200 µg/ml, respectively.

For drug susceptibility testing in the BACTEC 460TB system, final critical and high concentrations were 0.1 and 0.4 µg/ml, respectively, for isoniazid, 2.0 and 6.0 µg/ml, respectively, for streptomycin, and 2.5 and 7.5 µg/ml, respectively, for ethambutol. Rifampin and pyrazinamide were tested at critical concentrations of 2.0 µg/ml and 100 µg/ml, respectively. Centers 1 and 2 tested both critical and high concentrations for 81 susceptible strains. As no discrepancy was observed among these strains, centers 3 and 4 tested the higher concentrations only for strains showing resistance to any of the drugs at the critical concentration. Strains resistant at the critical concentration were considered low-level-resistance strains; strains resistant at both concentrations were classified as high-level-resistance strains.

Drug susceptibility testing. (i) MB/BACT system. We added 0.5 ml of the drug stock solution and 0.5 ml of the suspension containing *M. tuberculosis* to an MB/BACT bottle. The growth control did not contain any drugs. Drug susceptibility testing sets were entered into the MB/BACT instrument and continuously monitored until a positive or negative result was obtained. The drug susceptibility testing results were reported and interpreted with two methods (see below).

(ii) BACTEC 460TB system. Half a milliliter of a positive MB/BACT bottle was inoculated into a 12B vial and incubated till the growth index was ≥500. Drug susceptibility testing was done by following the standard procedure (15). Organisms initially grown on solid medium were inoculated in 12B vials and tested as soon as the growth index was ≥500.

Susceptibility testing interpretation. Two interpretative methods were used.

(i) Diluted control (10^{-1} control). An organism was determined to be susceptible when the antibiotic-containing bottle was not positive or had a positive time to detection greater than that of the 10^{-1} control. An organism was determined to be resistant when the antibiotic-containing bottle had a positive time to detection that was equal to or less than that of the 10^{-1} control.

(ii) Direct growth control. An organism was determined to be susceptible when the antibiotic-containing bottle was not detected as positive or had a positive time to detection greater than the sum of the time to detection for the direct control plus 3.5 days. An organism was determined to be resistant when the antibiotic-containing bottle had a positive time to detection that was less than or equal to the sum of the time to detection for the direct growth control plus 3.5 days.

The most accurate method in terms of sensitivity, specificity, and rapidity was adopted.

Reproducibility testing. Prior to testing clinical strains, a blind panel of nine strains of *M. tuberculosis* were sent to each center for reproducibility testing with the MB/BACT system by center 5. The expected results had been generated by center 5 with the reference method (proportion method on Löwenstein-Jensen slants). Centers 1, 3, and 4 tested the nine strains in duplicate at three cycles (thus, six replicates per strain). Center 2 tested eight strains in duplicate at two cycles (thus, four replicates per strain) and one strain in duplicate at one cycle. A total of 1,567 tests were realized (one vial was contaminated).

Quality control. A panel of seven reference strains of *M. tuberculosis* (ATCC 27294, ATCC 25618, ATCC 35822, ATCC 35838, ATCC 35820, ATCC 35837, and ATCC 35828) were sent by bioMerieux to the four centers. Quality control was performed at the beginning of the study and each time a new batch was introduced.

Resolution of discrepant results. Strains with discrepant results between the MB/BACT system and the BACTEC 460TB system were sent to center 5 for independent resolution by applying the proportion method on Löwenstein-Jensen slants. The bacterial suspension was adjusted to that of a McFarland no. 1. A 10^{-2} dilution of the bacterial suspension was then plated on Löwenstein-Jensen medium containing the desired concentrations of the drugs (isoniazid, 0.2, 0.5, 1, and 10 µg/ml; rifampin, 40 and 80 µg/ml; ethambutol, 2 and 4 µg/ml; streptomycin, 4 and 8 µg/ml; pyrazinamide, 200 µg/ml, pH 5.2). The slants were incubated at 37°C under a normal atmosphere. False-resistance results were defined as major errors, and false-susceptibility results were defined as very major errors. Performance parameters (sensitivity, specificity, positive predictive value, and negative predictive value) were determined after resolution of discrepant results.

TABLE 1. Drug susceptibility results for clinical strains of *M. tuberculosis* as determined by MB/BACT and BACTEC 460TB systems[a]

Drug (concn[b])	No. of tests	No. S by both tests	No. of results that were:		No. R by both tests	Agreement (%)
			R with MB/BACT, S with 460TB	S with MB/BACT, R with 460TB		
INH (0.09)	166	130		1	35	99.4
INH (0.4)	94	66			28	100
RIF (0.9)	166	148			18	100
EMB (3.5)	166	151		3	12	98.2
EMB (7.0)	82	76		4	2	95.1
STR (0.45)	166	139	2	1	24	98.2
STR (0.9)	85	70	1	3	11	95.3
PZA (200)	166	154		3	9	98.2
Total	1,091	934	3	15	139	98.3

[a] INH, isoniazid; RIF, rifampin; EMB, ethambutol; STR, streptomycin; PZA, pyrazinamide; S, susceptible; R, resistant.

[b] Concentrations are given in micrograms per milliliter.

RESULTS

As no significant difference was observed between the two controls, the method with the direct control was adopted as the most accurate one. The results obtained with the direct control were compared to those obtained by the proportion method. One thousand five hundred sixty-seven tests were realized with nine blind *M. tuberculosis* strains for testing the reproducibility of the MB/BACT system results. Full agreement of results was obtained for 1,509 tests (96.3%). Complete agreement between the MB/BACT system and the proportion method was found with isoniazid, rifampin, streptomycin, and ethambutol for more than 90% of strains. A slightly lower score obtained for pyrazinamide (84.2%) was explained by the lack of detection of a strain with low-level resistance to pyrazinamide.

One hundred sixty-six clinical strains of *M. tuberculosis* were tested for susceptibility to PRISE drugs at the critical (low) concentration. All the resistant and some susceptible strains at the critical concentration (see Materials and Methods, "Drug solutions") were tested at the higher concentrations of isoniazid (*n* = 94), ethambutol (*n* = 82), and streptomycin (*n* = 85) (Table 1). The isoniazid results agreed for 165 of 166 isolates tested at the critical concentration (99.4% agreement) and for the 94 strains tested at the higher concentration (100% agree-

ment). The rifampin results agreed for the 166 strains tested (100% agreement). The ethambutol results agreed for 163 of 166 strains at the critical concentration (98.2%) and for 78 of 82 at the higher concentration (95.1%). The streptomycin results agreed for 163 of the 166 strains tested at the critical concentration (98.2%) and for 81 of 85 isolates at the higher concentration (95.3%). For pyrazinamide, full agreement was found in 163 results out of the 166 strains tested (98.2%).

Fourteen strains were found to be discrepant between MB/BACT and BACTEC 460TB, 10 strains with one discrepant result and 4 strains with two discrepant results, amounting to 18 discrepant results out of a total of 1,091 tests (Table 1). Of those, 15 were susceptible according to MB/BACT but resistant according to BACTEC 460TB (isoniazid [*n* = 1], ethambutol [3 at the critical concentration, 4 at the higher concentration], streptomycin [1 at the critical concentration, 3 at the higher concentration], and pyrazinamide [*n* = 3]). All the discrepant strains were independent strains. Three strains were resistant to streptomycin according to MB/BACT but susceptible to streptomycin according to BACTEC 460TB (Table 1).

The resolution of discrepant results by the independent arbiter is presented in Table 2. The results of the proportion method agreed with the results generated by the MB/BACT

TABLE 2. Resolution of discrepant results with the proportion method on solid LJ medium[a]

Drug (concn[c])	No. of initial results		No. of resolved results[b]			
	R with MB/BACT, S with 460TB	S with MB/BACT, R with 460TB	R with MB/BACT and PM (true resistance)	S with MB/BACT and PM (true susceptibility)	R with MB/BACT, S PM (false-resistance ME)	S with MB/BACT, R PM (false-susceptibility VME)
INH (0.09)		1		1		
INH (0.4)						
RIF (0.9)						
EMB (3.5)		3		2		1
EMB (7)		4				4
STR (0.45)	2	1		1	2	
STR (0.9)	1	3	1			3
PZA (200)		3		3		
Total	3	15	1	7	2	8

[a] For an explanation of drug name abbreviations, see Table 1, footnote *a*. ME, major error; VME, very major error.

[b] Arbiter results based on the proportion method (PM).

[c] Concentrations are given in micrograms per milliliter.

TABLE 3. Accuracy of the MB/BACT compared with the
BACTEC 460TB system after resolution of discrepancies[a]

Drug (concn[b])	Sensitivity (%)	Specificity (%)	PPV (%)	NPV (%)
INH (0.09)	100	100	100	100
INH (0.4)	100	100	100	100
RIF (0.9)	100	100	100	100
EMB (3.5)	92.3	100	100	99.4
EMB (7.0)	33	100	100	98.3
STR (0.45)	100	98.6	92.3	100
STR (0.9)	80	100	100	99
PZA (200)	100	100	100	100

[a] For an explanation of drug name abbreviations, see Table 1, footnote a. PPV, positive predictive value; NPV, negative predictive value.
[b] Concentrations are given in micrograms per milliliter.

system in 44%, while 10 results remained discrepant for etham-butol and streptomycin. The accuracy of the MB/BACT system compared to that of the BACTEC 460TB system is presented in Table 3. The specificity (i.e., the ability to detect true susceptibility) was 100% for isoniazid, rifampin, ethambutol, pyrazinamide, and streptomycin at the high concentration and more than 98% for streptomycin at the critical concentration. Sensitivity (i.e., the ability to detect true resistance) was 100% for isoniazid, rifampin, and pyrazinamide. Sensitivity ranged from 80% to 100% for streptomycin and from 33% to 92.3% for ethambutol at the high and critical concentrations, respectively.

The RISE susceptibility test results with the MB/BACT system were obtained in 6.6 days on average (range, 6.0 to 9.9 days), whereas results were obtained in 5 days on average (range, 4.0 to 12.0 days) with the BACTEC 460TB system. The pyrazinamide susceptibility test results with the MB/BACT system were obtained in 7.8 days on average (range, 5.0 to 15.2 days), whereas results were obtained in 6.7 days on average (range, 3.0 to 13.0 days) with the BACTEC 460TB system.

DISCUSSION

The purpose of this multicenter study was to evaluate the reproducibility and reliability of the MB/BACT system for testing the susceptibility of *Mycobacterium tuberculosis* to the PRISE drugs. Most previous studies of the MB/BACT system have not included reproducibility testing (5, 6, 17, 20). In our study, both low- and high-level-resistance strains were tested for reproducibility. Good agreement was obtained for all the drugs at both concentrations and thus certified the reproducibility of the results. As no significant difference was observed between the two controls (data not shown), we suggest use of the direct control instead of the 10^{-1} one, which simplifies the workload and makes laboratory procedures safer.

Initial susceptibility testing yielded an overall good agreement of 98.3%. After the 18 discrepant cases were retested by an independent arbiter with the proportion method, there were eight falsely susceptible strains (very major errors), five with ethambutol and three with streptomycin, and two falsely resistant strains (major errors) with streptomycin by the MB/BACT system. Excellent agreement was obtained for the major antituberculous drugs isoniazid and rifampin (100% sensitivity and specificity). For rifampin, our agreement rates corroborate

those published earlier comparing the MB/BACT system with the agar proportion method (5, 6, 20). Comparing the MB/BACT system with the BACTEC 460TB system, Tortoli et al. (17) reported one major error among 113 *M. tuberculosis* strains tested with the MB/BACT system against rifampin. For isoniazid, our agreement rate was higher than those obtained in earlier studies with the MB/BACT system. Comparing the MB/BACT system with the agar proportion method, Brunello and Fontana (5) reported two very major errors out of 115 *M. tuberculosis* strains, and Yew et al. (20) found five major errors out of 105 *M. tuberculosis* strains with the MB/BACT system against isoniazid. Tortoli et al. (17) found one very major error with the MB/BACT system against isoniazid.

Earlier studies comparing the newly introduced BACTEC MGIT 960 system with the BACTEC 460TB system reported major errors with the BACTEC MGIT 960 system against isoniazid: Bemer et al. (2) found five major errors among 110 *M. tuberculosis* strains, and Tortoli et al. (17) found six major errors among 133 *M. tuberculosis* strains. Our data suggest the excellent ability of the MB/BACT system to detect true resistance and true susceptibility against isoniazid and rifampin, the two major front-line antituberculous drugs.

Among the first-line antituberculous drugs, ethambutol very often yields less reproducible results. A quality assurance program for drug susceptibility testing of *M. tuberculosis* was initiated in 1994 by the World Health Organization in 16 laboratories around the world (19). The first round of proficiency reported in 1997 yielded lower sensitivity values for ethambutol than for isoniazid and rifampin (66%, 99%, and 94%, respectively) (9). In the second round of proficiency reported in 2002 (10), the sensitivity of testing of ethambutol was less reliable, although it increased from 60% in round 1 to 98% in round 5. As a consequence, the sensitivity of ethambutol leads to underreporting of drug resistance.

Using the MB/BACT system, Brunello and Fontana (5) found five major errors among 115 strains, Diaz-Infantes et al. (6) found three very major errors and two major errors among 83 strains, and Tortoli et al. (17) found three very major errors and two major errors among 113 strains against ethambutol. The three studies used a critical concentration of ethambutol of 2 μg/ml. The manufacturer decided to increase the final critical concentration of ethambutol to 3.5 μg/ml. At this concentration, tested in our study, there were no false-resistance results without an increase in the false-susceptibility results (one very major error) by comparison with the previous studies. At the high concentration of 7.0 μg/ml, four very major errors were found. It was remarkable that the four falsely susceptible strains were found to be truely resistant at the critical concentration.

Of the five discrepancies observed with streptomycin, there were two major errors at the critical concentration and three very major errors at the high concentration with the MB/BACT system. Nevertheless, the MB/BACT system is reliable in detecting truely resistant strains at the critical concentration, as the three very major errors at the high concentration were found to indicate true resistance at the critical concentration.

Out of the eight very major errors found with the MB/BACT system, seven were obtained at high concentrations. This observation suggests that the high concentrations of streptomycin (0.9 μg/ml) and ethambutol (7.0 μg/ml) might be too high.

While this may hold true for ethambutol, it certainly does not for streptomycin, since false-susceptibility results were reported at 1.0 μg/ml (6, 17, 20) but not at 2.0 μg/ml (5) with the MB/BACT system. In fact, the need for testing high concentrations of ethambutol and streptomycin is not clearly defined.

Our study is the first study with a standardized kit for susceptibility testing of *M. tuberculosis* to pyrazinamide with the MB/BACT system. An excellent agreement was obtained for pyrazinamide (100% sensitivity, specificity, positive predictive value, and negative predictive value). Previous evaluations of newer antimycobacterial susceptibility testing systems, the ESP and the BACTEC MGIT 960 systems, found discrepant results when testing pyrazinamide susceptibility among *M. tuberculosis* isolates (1, 8, 12). Comparing the ESP system with the BACTEC 460TB system, LaBombardi (8) reported one very major error and one major error among 50 *M. tuberculosis* strains tested with the BACTEC MGIT 960 system. Comparing the BACTEC MGIT 960 system with the BACTEC 460TB system, Pfyffer et al. (12) reported one very major error and three major errors among 116 *M. tuberculosis* strains tested with the BACTEC MGIT 960 system. The absence of any false-susceptibility or false-resistance results with MB/BACT indicates the excellent ability of the system for rapid testing of the susceptibility of *M. tuberculosis* to pyrazinamide.

The median time for obtaining RISE susceptibility results was 6.6 days, which is similar to that obtained by the BACTEC 460TB system (5.0 days) and shorter than that observed by Brunello and Fontana (5) and Tortoli et al. (17) with the MB/BACT system (8.5 and 11.6 days, respectively). The times required for RISE susceptibility testing of *M. tuberculosis* did not differ between the direct and 10^{-1} controls. The median time for obtaining pyrazinamide susceptibility results was 7.8 days, which is similar to that obtained by the BACTEC 460TB system (6.7 days). The mean turnaround times for pyrazinamide susceptibility testing with the MB/BACT system were similar to those obtained by Aono et al. (1) and Pfyffer et al. (12) with the BACTEC MGIT 960 system (7.7 and 6.8 days, respectively). Nevertheless, 1 day more was required to achieve the final pyrazinamide results with the 10^{-1} control (median time, 9 days), with a very wide range (4.0 to 36.5 days) (results not shown). Some isolates of *M. tuberculosis* failed to grow in an acidic medium, especially for diluted mycobacterial suspensions, which can explain the very long time (36.5 days) required for pyrazinamide susceptibility testing of some *M. tuberculosis* isolates with the proportional control.

In summary, our study demonstrates that (i) the MB/BACT system is a reliable method for testing the susceptibility of *M. tuberculosis*; (ii) the overall agreement of results is excellent for the three major antituberculous drugs, isoniazid, rifampin, and pyrazinamide; (iii) additional studies are required in order to improve ethambutol and streptomycin testing results, particularly at the high concentration; (iv) an undiluted growth control (the direct control) should be used, especially for testing pyrazinamide; and (v) the MB/BACT turnaround time for PRISE testing is as fast as that of the radiometric method.

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