

Evaluation of the MicroScan Antimicrobial Susceptibility System with the autoSCAN-4 Automated Reader

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The American MicroScan (American MicroScan, Mahwah, N.J.) identification and antimicrobial susceptibility system consists in part of an automated reading system (autoSCAN-4) with data management capabilities. We evaluated the system with 404 gram-negative and 170 gram-positive facultative anaerobic and aerobic bacteria. We compared MicroScan results read automatically and visually with each other and with the results obtained by the reference method (read visually). The overall agreement within $\pm 1 \log_2$ dilution was 94.3% when the MicroScan system (read automatically) was compared with the reference method (read visually), 96.4% when MicroScan panels (read visually) were compared with reference panels, and 97.4% when the autoSCAN-4 automated reading was compared with the visual reading of the MicroScan panels. Total discrepancies (susceptibility interpretation category changes) for the MicroScan system compared with the reference method were 7%, with 6.2% considered a minor discrepancy. The autoSCAN-4 and the complete MicroScan system yielded accurate results compared with the reference method.

Increased use of broth microdilution for performing antimicrobial susceptibility tests has led to the development of automated readers and data management systems in the microbiology laboratory. One microdilution system is the MicroScan (American MicroScan, Mahwah, N.J.) which uses commercial trays that can be read visually, but which also may contain an automated reader, the autoSCAN-4. The system is also capable of identifying bacteria and has a data management system.

In this study we evaluated the accuracy of the MicroScan system by comparing the results of automated readings with those of manual readings and by comparing each of these results with those obtained with a reference microdilution system. We challenged these systems with organisms selected to have diverse and unusual susceptibility patterns, known mechanisms of resistance, and some with a history of causing problems when tested for antimicrobial susceptibility.

MATERIALS AND METHODS

We tested 574 bacterial cultures for this study. They included 306 isolates of the family *Enterobacteriaceae* (28 *Citrobacter* spp., 6 *Edwardsiella* spp., 40 *Enterobacter* spp., 28 *Escherichia* spp., 10 *Hafnia* spp., 31 *Klebsiella* spp., 1 *Kluyvera* spp., 10 *Morganella* spp., 20 *Proteus* spp., 35 *Providencia* spp., 37 *Salmonella* spp., 29 *Serratia* spp., 22 *Shigella* spp., and 9 *Yersinia* spp.); 69 nonfermentative species (5 *Archromobacter* spp.; 10 *Acinetobacter* spp.; 2 *Alcaligenes* spp.; 3 *Bordetella* spp.; 7 cultures of CDC groups: 1 Iif, 3 IVc-2, 2 Ve-1, 1 Ve-2; 6 *Moraxella* spp.; and 36 *Pseudomonas* spp.); and 29 fermentative isolates other than the family *Enterobacteriaceae* (2 *Aeromonas* spp., 4 *Chromobacterium* spp., 5 *Flavobacterium* spp., 9 *Pasteurella* spp., and 9 *Vibrio* spp.). We also tested 170 gram-positive bacterial isolates (40 *Staphylococcus aureus*: 19 methicillin susceptible and 21 methicillin resistant; 27 coagulase-negative *Staphylococcus* spp.; 86 *Streptococcus* spp.; 19

alpha-hemolytic, 29 beta-hemolytic, 24 enterococcus, and 14 pneumococcus; 7 *Corynebacterium* spp.; 3 *Bacillus* spp.; 2 *Lactobacillus* spp.; 3 *Listeria* spp.; 1 *Rhodococcus* sp.; and an unclassified group [1 gram-positive rod]). These organisms grew adequately in the MicroScan medium and in the reference medium for comparison of susceptibility test data.

The following organisms, however, did not have sufficient growth for susceptibility testing by either method: one *S. aureus*, one *Staphylococcus epidermidis*, two *Corynebacterium* spp., one *Lactobacillus* sp., three *Bordetella* spp., and four *Pasteurella* spp. Some other strains had insufficient growth in one or the other of the systems; for the MicroScan system, this included four *Streptococcus pneumoniae*, and for the reference method, five *Corynebacterium* species had insufficient growth. These organisms were excluded when the data were analyzed and were not included in the total number tested.

The isolates were obtained from either our stock culture collection or from the stock culture collection of the Respiratory and Special Pathogens Branch of the Centers for Disease Control. The organisms were removed from storage, where they had been frozen in blood or held in motility medium at room temperature for an indefinite period, and subcultured two times on Trypticase soy agar (BBL Microbiology Systems, Cockeysville, Md.) containing 5% sheep blood. The inoculum used for susceptibility testing was prepared by suspending growth taken from the Trypticase soy agar in 5 ml of Mueller-Hinton broth and adjusting the inoculum to a turbidity equal to a 0.5 McFarland standard (approximately 10^8 CFU/ml). This adjusted inoculum was used for both the reference and MicroScan methods. The microdilution trays for the reference method were prepared in-house with 12 or 15 antimicrobial agents each for the gram-positive and gram-negative bacteria, respectively. The antimicrobial concentrations used were the same as those used in the MicroScan system. The antimicrobial powders were kindly supplied by the respective manufacturers. The reference broth microdilution susceptibility trays were prepared and tested as described in the National Committee for

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TABLE 1. Percent overall agreement of MICs of 15 antimicrobial agents for 404 gram-negative bacteria when MicroScan visual readings were compared with reference method visual readings (6,060 tests)

Antimicrobial agent	% Agreement at indicated dilution difference (no. of tests) ^a						
	>-2 (3)	-2 (56)	-1 (588)	0 (4,963)	+1 (377)	+2 (58)	>+2 (15)
Ampicillin	0.7	8.2	31.2	56.9	2.5	0.5	0
Carbenicillin	0	0	11.4	77.0	7.9	3.0	0.7
Piperacillin	0	0.5	1.5	91.3	4.7	0.7	1.3
Cephalothin	0	0.5	10.9	80.2	7.2	0.7	0.5
Cefamandole	0	1.7	5.4	85.4	5.5	2.0	0
Cefoxitin	0	1.0	13.9	75.5	8.1	1.5	0
Cefoperazone	0	0	5.0	92.1	2.7	0.2	0
Cefotaxime	0	0	1.2	95.6	3.2	0	0
Moxalactam	0	0	3.2	94.3	1.7	0.5	0.3
Amikacin	0	0.2	8.2	74.5	14.6	2.0	0.5
Gentamicin	0	0.2	4.0	86.1	8.2	1.5	0
Tobramycin	0	0	7.9	83.7	7.4	1.0	0
Chloramphenicol	0	0.5	29.2	66.6	3.0	0.5	0.2
Tetracycline	0	0.5	10.4	73.8	15.1	0	0.2
Sulfamethoxazole/ trimethoprim ^b	0	0.5	2.2	95.6	1.5	0.2	0

^a 0 indicates that results were the same, +1 that the MicroScan reading was 1 log₂ dilution higher, etc.

^b 19 parts sulfamethoxazole plus 1 part trimethoprim.

Clinical Laboratory Standards (NCCLS) tentative standard for dilution testing M7-T (9).

The MicroScan system microdilution trays were prepared and supplied frozen by American MicroScan. Gram-negative bacteria were tested in the negative combo panel, which contains both diluted antimicrobial and biochemical agents for identification; gram-positive bacteria were tested in the positive MIC panels, which contain only diluted antimicrobial agents. Tests were performed according to the written directions supplied by the manufacturer.

MICs were read as the lowest concentration of antimicrobial agent that inhibited visible growth of the bacteria. Reference trays were read only visually, but the MicroScan trays were read visually and automatically by the autoSCAN-4 reader. All results were recorded and stored on the IBM personal computer, which is a component of the MicroScan system.

Each well of the test microdilution plate was read by detecting the amount of light that passed through it at wavelengths of 620, 560, 505, 470, 440, and 590 nm. These

transmitted light values were converted to analogs, and the results were interpreted by the computer. All 96 wells of the microdilution trays were read, and calculations were made in less than 30 s, with the actual reading time being approximately 6 s.

RESULTS

The results of this study were analyzed in three ways. (i) Results from the MicroScan method read visually were compared with results from the reference method read visually. (ii) Results from the MicroScan method read by the autoSCAN-4 were compared with results from the reference method read visually. (iii) Results from the MicroScan method read by the autoSCAN-4 were compared with the results from the MicroScan method read visually.

The overall percent agreement for the MICs obtained of the antimicrobial agents tested for gram-positive and gram-negative bacteria is shown in Tables 1 through 6. The MicroScan method versus the reference method (Table 1) had 97.8% agreement within ± 1 log₂ dilution with gram-

TABLE 2. Percent overall agreement of MICs of 12 antimicrobial agents for 170 gram-positive bacteria when MicroScan visual readings were compared with reference method visual readings (2,040 tests)

Antimicrobial agent	% Agreement at indicated dilution difference (no. of tests) ^a						
	>-2 (45)	-2 (90)	-1 (302)	0 (1,479)	+1 (102)	+2 (14)	>+2 (8)
Ampicillin	2.4	8.2	22.9	64.1	2.4	0	0
Nafcillin	2.9	2.4	11.8	60.0	12.4	7.0	3.5
Penicillin	1.7	6.5	18.2	70.6	2.4	0.6	0
Cephalothin	2.4	4.1	10.6	80.0	2.9	0	0
Chloramphenicol	0	7.0	60.0	32.4	0.6	0	0
Clindamycin	0	0	2.9	94.2	2.9	0	0
Erythromycin	0.6	0	9.4	86.5	2.9	0.6	0
Gentamicin	0	0	1.2	72.3	26.5	0	0
Vancomycin	0	0.6	8.8	88.2	2.4	0	0
Nitrofurantoin	0	0	12.9	83.0	4.1	0	0
Tetracycline	16.5	24.1	18.8	38.6	0.6	0	1.2
Sulfamethoxazole/ Trimethoprim ^b	0	0	0	100.0	0	0	0

^a See Table 1, footnote *a*.

^b See Table 1, footnote *b*.

TABLE 3. Percent overall agreement of MICs of 15 antimicrobial agents for 404 gram-negative bacteria when results from the MicroScan method read by the autoSCAN-4 were compared with reference method visual readings (6,060 tests)

Antimicrobial agent	% Agreement at indicated dilution difference (no. of tests) ^a						
	>-2 (21)	-2 (140)	-1 (889)	0 (4,608)	+1 (277)	+2 (103)	>+2 (22)
Ampicillin	1.0	5.9	23.5	67.9	1.0	0.7	0
Carbenicillin	0	0.3	13.6	67.1	12.9	5.9	0.2
Piperacillin	0	0	2.3	87.4	6.9	1.7	1.7
Cephalothin	0.3	2.5	18.3	73.5	3.7	1.2	0.5
Cefamandole	0.7	1.5	9.7	80.9	3.5	3.5	0.2
Cefoxitin	0	2.2	24.8	65.3	6.2	0.5	1.0
Cefoperazone	0	0	6.2	89.3	4.0	0.5	0
Cefotaxime	0	0	3.2	93.8	2.7	0.3	0
Moxalactam	0	0.5	6.7	86.4	5.9	0.5	0
Amikacin	0.3	1.5	12.6	71.0	8.9	5.7	0
Gentamicin	0	0.5	4.5	86.6	5.0	2.5	0.9
Tobramycin	0	0.5	11.4	84.2	3.0	0.7	0.2
Chloramphenicol	1.5	10.6	39.4	45.5	2.0	0.5	0.5
Tetracycline	1.5	8.2	38.1	50.0	1.7	0.5	0
Sulfamethoxazole/ trimethoprim ^b	0	0.5	5.9	91.6	1.2	0.8	0

^a See Table 1, footnote a.

^b See Table 1, footnote b.

negative bacteria. There was 92.3% agreement with the two methods for gram-positive bacteria (Table 2). The MicroScan method read by the autoSCAN-4 agreed within $\pm 1 \log_2$ dilution with the reference method at 95.3% for gram-negative bacteria (Table 3) and 91.5% for gram-positive bacteria (Table 4). The MicroScan method read by the autoSCAN-4 compared with MicroScan readings had 96.8% agreement to within $\pm 1 \log_2$ dilution for gram-negative bacteria (Table 5) and had 99.0% agreement for gram-positive bacteria (Table 6).

If the data are analyzed for overall agreement within $\pm 2 \log_2$ dilutions, the values for the MicroScan versus the reference method were 99.7 and 97.4%, for the MicroScan method read by the autoSCAN-4 versus the reference method the values were 99.3 and 97.6%, and for the MicroScan method read by the autoSCAN-4 versus the

MicroScan method the results were 99.3 and 99.3% for gram-negative and gram-positive bacteria, respectively.

For gram-negative bacilli, the MicroScan system tended to yield lower MICs than the reference method, whether the results were read visually or automatically. The only drugs that yielded >10% of the MICs that were one dilution higher than the reference MICs (Tables 1 and 3) were amikacin (14.6%) and tetracycline (15.1%) when the MicroScan was read visually and carbenicillin (12.9%) when it was read automatically. On the other hand, ampicillin, carbenicillin, cephalothin, cefoxitin, chloramphenicol, and tetracycline had >10% MICs that were one dilution lower when the visually read MicroScan results were compared with the reference results; these six antimicrobial agents plus amika-

TABLE 4. Percent overall agreement of MICs of 12 antimicrobial agents for 170 gram-positive bacteria when results from the MicroScan read by the autoSCAN-4 were compared with reference method visual readings (2,040 tests)

Antimicrobial agent	% Agreement at indicated dilution difference (no. of tests) ^a						
	>-2 (43)	-2 (113)	-1 (328)	0 (1,463)	+1 (75)	+2 (13)	>+2 (5)
Ampicillin	3.5	6.5	25.9	62.4	1.7	0	0
Nafcillin	5.9	2.4	11.7	64.1	10.0	5.3	0.6
Penicillin	2.4	7.0	18.8	70.6	0.6	0.6	0
Cephalothin	2.9	4.7	11.2	78.3	2.9	0	0
Chloramphenicol	0	10.0	65.9	23.5	0.6	0	0
Clindamycin	0	1.2	2.9	95.3	0.6	0	0
Erythromycin	0.6	1.2	10.0	85.9	1.7	0	0.6
Gentamicin	0	0	4.1	75.9	19.4	0.6	0
Vancomycin	0	0	9.4	89.4	1.2	0	0
Nitrofurantoin	0	0.6	14.7	77.6	5.3	1.2	0.6
Tetracycline	10.0	32.9	18.2	37.7	0	0	1.2
Sulfamethoxazole/ trimethoprim ^b	0	0	0	100.0	0	0	0

^a See Table 1, footnote a.

^b See Table 1, footnote b.

TABLE 5. Percent overall agreement of MICs of 15 antimicrobial agents for 404 gram-negative bacteria when results from the MicroScan read by the autoSCAN-4 were compared with MicroScan method visual readings (6,060 tests)

Antimicrobial agent	% Agreement at indicated dilution difference (no. of tests) ^a						
	>-2 (11)	-2 (42)	-1 (594)	0 (5,080)	+1 (192)	+2 (107)	>+2 (34)
Ampicillin	1.0	2.0	14.1	80.4	1.2	1.0	0.3
Carbenicillin	0	0.7	5.2	84.7	7.4	1.7	0.3
Piperacillin	0.5	0.2	3.0	87.4	5.2	1.7	2.0
Cephalothin	0.3	1.2	13.4	82.2	1.7	0.7	0.5
Cefamandole	0.2	0.3	7.2	84.4	1.7	5.2	1.0
Cefoxitin	0.5	0.2	15.1	79.0	2.2	1.5	1.5
Cefoperazone	0	0.3	1.5	94.3	2.7	1.2	0
Cefotaxime	0	0	2.0	96.3	1.5	0.2	0
Moxalactam	0	0.5	2.2	89.1	7.2	1.0	0
Amikacin	0.2	0.5	9.9	82.2	3.0	3.7	0.5
Gentamicin	0	0.2	2.7	84.2	7.9	3.5	1.5
Tobramycin	0	0.5	6.7	89.4	1.2	2.0	0.2
Chloramphenicol	0	2.0	23.3	70.6	2.7	0.7	0.7
Tetracycline	0	1.7	36.1	59.4	1.0	1.8	0
Sulfamethoxazole/ trimethoprim ^b	0	0	4.7	94.1	0.7	0.5	0

^a See Table 1, footnote a.

^b See Table 1, footnote b.

TABLE 6. Percent overall agreement of MICs of 12 antimicrobial agents for 170 gram-positive bacteria when results from the MicroScan read by the autoSCAN-4 were compared with MicroScan method visual readings (2,040 tests)

Antimicrobial agent	% Agreement at indicated dilution difference (no. of tests) ^a						
	>-2 (15)	-2 (3)	-1 (80)	0 (1,918)	+1 (23)	+2 (1)	>+2 (0)
Ampicillin	0.6	0.6	0.6	97.0	1.2	0	0
Nafcillin	5.3	1.2	10.6	81.7	1.2	0	0
Penicillin	0.6	0	3.5	95.9	0	0	0
Cephalothin	0.6	0	1.8	97.6	0	0	0
Chloramphenicol	0	0	12.9	84.7	2.4	0	0
Clindamycin	0.6	0	2.9	95.9	0.6	0	0
Erythromycin	1.2	0	1.2	96.4	1.2	0	0
Gentamicin	0	0	8.2	91.8	0	0	0
Vancomycin	0	0	0.6	98.2	1.2	0	0
Nitrofurantoin	0	0	1.2	93.5	4.7	0.6	0
Tetracycline	0	0	3.5	95.3	1.2	0	0
Sulfamethoxazole/ trimethoprim ^b	0	0	0	100.0	0	0	0

^a See Table 1, footnote a.

^b See Table 1, footnote b.

cin and tobramycin had >10% of MICs that were one dilution lower when automated results were compared with the reference results. At the ± 2 dilution level, only ampicillin, carbenicillin, chloramphenicol, and tetracycline showed substantial numbers of errors (Tables 1 and 3).

Automated MicroScan results most often yielded the same MICs as did the MicroScan read visually. When the results differed, however, the automated results showed some tendency to read one dilution lower than the visual results; this occurred more often with chloramphenicol and tetracycline.

For gram-positive bacteria, the MicroScan results also tended to be lower than those of the reference method. The antimicrobial agents showing these tendencies were ampicillin, penicillin, cephalothin, chloramphenicol, erythromycin, vancomycin, nitrofurantoin, and tetracycline. Gentamicin results from the MicroScan system tended to be one dilution higher than results with the reference method, while nafcillin was about equally split between +1 dilution and -1 dilution. The number of discrepancies was greater with gram-positive organisms than with gram-negative organisms, as was shown by the greater number of MicroScan results that were at least two dilutions lower than results from the reference method (Tables 1 through 4). The most errors were seen with tetracycline in tests with gram-positive bacteria; approximately 40% of the MicroScan results were at least two dilutions lower than results with the reference method.

The MICs obtained by each method were placed into the categories of susceptibility defined in the NCCLS document M7-T (9). Discrepancies in the categories obtained by MicroScan (visual and automated readings) compared with those obtained with the reference method were defined as previously described: very major (susceptible by MicroScan and resistant by the reference method), major (resistant by MicroScan and susceptible by the reference method), or minor (one of the results was intermediate) (11). Most of the errors in categorization were minor (Table 7). Bacterial species for which minor discrepancies were >10% for an antimicrobial agent are shown in Table 8. The largest number of discrepancies was obtained with ampicillin and tetracycline, which occurred with most species.

There were no major discrepancies >5% for any antimicrobial agent-organism combination. In addition, when all

gram-negative bacteria are examined, no antimicrobial agent had >1% very major discrepancies. If, however, *Serratia* species alone are examined, cefoxitin had 10% (3 of 30) very major discrepancies and 23% (7 of 30) minor discrepancies. Also, 4 of 4 *Proteus mirabilis* strains resistant to ampicillin had minor discrepancies, but none of the ampicillin-susceptible *P. mirabilis* strains were discrepant with ampicillin. *Salmonella* species had a large number of minor discrepancies with tetracycline (15 of 37) and with ampicillin (17 of 37).

There were, however, gram-positive bacteria-antimicrobial agent combinations with >1% very major discrepancies (Table 9). The largest number of these very major discrepancies occurred with cephalothin and *S. aureus*, but nafcillin discrepancies were found in several species.

Discrepancies because of the automated reader were only occasionally seen. In some cases, these were caused by bubbles trapped in the medium or by scratches or flaws in the plastic wells of the microdilution trays.

DISCUSSION

The efficacy of commercial microdilution systems in determining the MICs of antimicrobial agents for clinically important bacteria has been established (1, 2, 5-8, 10, 12). On the other hand, systems offering automated readings of the antimicrobial microdilution susceptibility test represent more recent steps toward the automation or mechanization of the microbiology laboratory and have been reported less often (3, 4, 8). We evaluated the MicroScan susceptibility testing system for accuracy of the method, i.e., its accuracy if read visually, and for the accuracy of the autoSCAN-4 automated reader. The results yielded by the MicroScan antimicrobial microdilution system were acceptable, whether they were read visually or with the automated reader, especially for gram-negative bacteria.

The MicroScan gram-positive MIC panel uses a modified broth for testing gram-positive organisms, whereas the reference method uses Mueller-Hinton broth. This difference in basal medium for the two methods may partially account for the lower correlation of the test panels with the gram-positive organisms. In fact, as noted above, some gram-positive organisms grew in one medium but not in the other and thus could not be evaluated. Different amounts of growth in the two systems may have accounted for the differences seen with the β -lactam antimicrobial agents and the bacteriostatic antimicrobial agents. Most of the gram-positive organisms that had ± 2 log₂ dilution differences or more were *Corynebacterium* species, methicillin-resistant *S. aureus*, and *S. pneumoniae*. All of these organisms can be difficult to test accurately, and all of the discrepancies were

TABLE 7. Overall category changes for results obtained with the MicroScan method read by the autoSCAN-4 compared with the reference method

Bacteria	% Discrepancy with indicated category of susceptibility ^a		
	Minor (6.21) ^b	Major (0.48) ^b	Very major (0.38) ^b
Gram-positive	5.7	0.5	1.3
Gram-negative	6.4	0.4	0.08

^a Minor, one result was intermediate; major, susceptible by the reference method but resistant by the MicroScan method; very major, susceptible by the MicroScan method but resistant by the reference method.

^b Total percent in each category.

TABLE 8. Minor discrepancies (>10%) with specific antimicrobial agent-bacterial species combinations when results from the MicroScan method read by the autoSCAN 4 were compared with results from the reference method

Bacteria (no. tested)	% Strains showing >10% minor discrepancies with indicated antimicrobial agent (total %)				
	Ampicillin (16.4)	Carbenicillin (10.6)	Piperacillin (10.4)	Gentamicin (3.8)	Tetracycline (18.1)
Gram negative (404)	18	10.6	10.4	0	18
<i>Acinetobacter</i> spp.					X ^a
<i>Citrobacter</i> spp.	X	X			X
<i>Enterobacter</i> spp.	X	X	X		X
<i>Escherichia</i> spp.	X				X
<i>Klebsiella</i> spp.	X	X			X
<i>Proteus</i> spp.	X	X			X
<i>Providencia</i> spp.	X				X
<i>Pseudomonas</i> spp.		X			
<i>Salmonella</i> spp.	X				X
<i>Serratia</i> spp.	X	X			
<i>Shigella</i> spp.	X	X			X
Miscellaneous nonfermenting			X		
Gram positive (170)	12.4	ND ^b	ND	12.9	18.2
<i>Staphylococcus aureus</i>	X				X
Coagulase-negative staphylococci					X
<i>Streptococcus pneumoniae</i>	X			X	
β-Streptococci				X	
Enterococci	X				
Miscellaneous rods	X				X

^a Combination in which discrepancies occurred.
^b ND, Not done.

with the β-lactam antimicrobial agents. Although there were few discrepancies with the gram-negative bacteria, most of the discrepancies that did occur were not surprising, since they had been reported for other systems (6, 7). For example, Jones et al. (7) reported that problem combinations were *Acinetobacter calcoaceticus* subsp. *anitratus*- and *Proteus vulgaris*-ampicillin and *Enterobacter aerogenes*-cephalothin; in another study they reported problems with *Enterobacter cloacae*- and *Providencia rettgeri*-ampicillin, *Klebsiella pneumoniae*- and *Klebsiella oxytoca*-carbenicillin, and *Escherichia coli*-cephalothin (6).

TABLE 9. Gram-positive bacteria-antimicrobial agent combinations with >1% very major discrepancies when results from the MicroScan read by the autoSCAN 4 were compared with results from the reference method

Bacteria (no., 170)	% Strains showing >1% very major discrepancies with indicated antimicrobial agent (total %)				
	Cephalothin (5.9)	Nafcillin (4.7)	Penicillin (3.5)	Tetracycline (1.8)	Nitrofurantion (2.4)
<i>Staphylococcus aureus</i>	X ^a	X	X		
Coagulase-negative staphylococci			X	X	
<i>Streptococcus pneumoniae</i>		X ^b			
β-streptococci					X
Enterococci		X ^b			
Miscellaneous ^c		X ^b		X	X

^a Combination in which discrepancies occurred.
^b Nafcillin results would be reported by the MicroScan method only for *Staphylococcus* species.
^c Includes *Corynebacterium*, *Lactobacillus*, α-streptococci, and *Rhodococcus* species.

The autoSCAN-4 readings compared with laboratory personnel readings of the same tray were acceptable in that they had 96.8% agreement within ±1 log₂ dilution for gram-negative bacteria and 99.0% agreement for gram-positive bacteria. When there was a difference between visual readings and autoSCAN-4 readings, the autoSCAN-4 tended to be 1 log₂ dilution lower than the visual reading. We do not know why this tendency to lower MICs occurs in the commercial system, but it may be due to the difficulty of the optical systems in reading results with some antimicrobial agents, e.g., chloramphenicol and tetracycline. But the rare differences of greater than or equal to 2 log₂ dilutions were usually caused by skipped wells, debris, bubbles, or scratches in the wells of the microdilution trays. Tetracycline and chloramphenicol with gram-negative bacteria and nafcillin and chloramphenicol with gram-positive bacteria were the antimicrobial agents that were the most difficult for the autoSCAN-4 to read; they were more frequently read with lower results than any of the other antimicrobial agents. The problem with nafcillin did not occur with *Staphylococcus* species, which usually read the same, but with *Streptococcus* species in which MICs of nafcillin for the organism were lower. Based on these data, we concluded that the automated reader was acceptably accurate.

As indicated above, we analyzed the data by looking at interpretation or category changes using the M7-T guidelines (9) to determine the category of susceptibility. We found most discrepancies to be minor. Many of the minor discrepancies that occurred with both gram-positive and gram-negative bacteria for ampicillin and tetracycline could be attributed to the category breakpoints used in the M7-T standard. For an MIC of 1 μg/ml, the organism would be judged susceptible, but if it were 2 μg/ml it would be judged moderately susceptible. Therefore, for those organisms inhibited by 1 or 2 μg of either antimicrobial agent per ml, a random 1 dilution change in MIC would cause a change in

the category interpretation of susceptibility. Despite this, the overall minor discrepancies were still only 6.2%, which is within the acceptable level of disagreement suggested by Thornsberry and Gavan (11) when a new system is tested. Major and very major discrepancies for the system were less than 5%, which is the guideline suggested by Thornsberry and Gavan (11).

We believe that the evaluation of a new system should include not only the organisms isolated in an average clinical laboratory but also a challenge set of bacteria, which have diverse and unusual susceptibility patterns and include known resistance mechanisms and organisms that are known to cause problems when performing *in vitro* antimicrobial susceptibility tests. The organisms used in this study are from such a challenge set. They probably offer a greater challenge to the system than the usual set of clinical isolates, since organisms in the general population would be unlikely to contain the problem bacteria and would usually give fewer discrepancies than those examined in this study. Thus the MicroScan system performed accurately and reliably under more stringent conditions than might be found in a clinical laboratory.

In conclusion, the MicroScan system with its automated reader yields MICs that are accurate and reproducible and can be read relatively rapidly. Whenever differences occurred, the MicroScan system tended to yield MICs that were one dilution lower than those obtained with a reference method.

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