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Comparison and Evaluation of Osiris and Sirscan 2000 Antimicrobial Susceptibility Systems in the Clinical Microbiology Laboratory

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The Osiris and Sirscan 2000 systems are two semiautomated systems that can be used to read and interpret the results on disk diffusion agar plates. They are both used for determination of susceptibility to antimicrobial agents. The present study compared both systems versus the NCCLS standard method of visual reading with a ruler. Both inpatient and outpatient samples with a total of 315 nonfastidious gram-negative strains were obtained. In total, 3,724 organism-antimicrobial agent combinations that fulfilled the NCCLS guidelines for disk diffusion susceptibility testing were evaluated prospectively. The results obtained with both systems in comparison with those obtained by the classical nonautomated means of interpretation were excellent, with correlation coefficients of 0.96 for both systems. The overall agreements for susceptibility interpretation were 96.56 and 96.24% with the Osiris and Sirscan systems, respectively. Very major errors were obtained for 8 (1.07%) and 10 (1.34%) organism-antimicrobial agent combinations with the Osiris and Sirscan systems, respectively. In addition, major errors were obtained for 2 (0.07%) and 6 (0.21%) combinations with the Osiris and Sirscan systems, respectively. Minor errors were obtained for 118 and 124 organism-antimicrobial agent combinations with the Osiris and Sirscan systems, respectively. Overall, both the Osiris system and the Sirscan system are comparable and reliable systems for determination of interpretative categories from the zone diameters of standard disk diffusion test plates.

An important task for the diagnostic clinical microbiology laboratory is the detection of clinically relevant antimicrobial resistance in individual isolates (4). Many different techniques are available for the testing of susceptibility, including the disk diffusion (Kirby Bauer), broth microdilution, agar dilution, and antibiotic gradient methods (5). A variety of automated instruments based on these different methodologies have been introduced. The potential advantages of automation include standardization, which results in increased accuracy; the faster availability of results; improved data management; and the possibility for the use of artificial intelligence.

Kirby Bauer disk diffusion susceptibility testing merely provides susceptibility category results (susceptible, intermediate, and resistant). On the other hand, measurement of zone sizes is tedious, time-consuming, and fraught with transcription errors. Disk diffusion nevertheless offers certain advantages over other methods: low cost, the ability to test large numbers of organisms, and the ease with which different antimicrobial agents can be chosen (2). In addition, there is no evidence that determination of MICs is superior to determination of susceptibility category in patient management (4).

The Osiris system (Sanofi Pasteur) and the Sirscan 2000 system (i2a, Montpellier, France) are two semiautomated data management systems developed to combine the advantages of automation and disk diffusion. These video systems measure the inhibition zone diameter and interpret the results for disk diffusion susceptibility agar plates. The agar plate is placed onto a sliding tray, and the reading of the zone diameter is initiated by a keystroke. A clear image with fully calculated zone diameters appears on the video screen. Both manufacturers recommend a review of each plate by an experienced technologist so that adjustments can be made, if necessary. A user-programmed expert system screens the results for each isolate.

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MATERIALS AND METHODS

Three hundred fifteen gram-negative aerobic bacilli of 9 genera and 12 species were included in this study (Table 1). In total, 3,724 organism-antimicrobial agent combinations were tested prospectively. The organisms were isolated from inpatients and outpatients at our hospital. The organisms tested are representative of the rapidly growing nonfastidious gram-negative aerobe organism mix recovered at our hospital. The isolates were obtained from different sources: urine samples (41%), sputum and tracheal aspirates (34%), blood (3%), and miscellaneous sources. The antimicrobial agents tested depended on both the identification and the source of the organism. Members of the family *Enterobacteriaceae* isolated from urine were tested with amikacin, amoxicillin-clavulanic acid, ampicillin, aztreonam, cefazolin, cefotaxime, ceftazidime, cefuroxime, netilmicin, nitrofurantoin, norfloxacin, piperacillin, and trimethoprim-sulfamethoxazole. *Escherichia coli* strains obtained from urine were also tested with fosfomycin. *Pseudomonas aeruginosa* and *Acinetobacter* sp. strains isolated from urine were tested with amikacin, aztreonam, ceftazidime, netilmicin, norfloxacin, and piperacillin. Members of the family *Enterobacteriaceae* from sites other than urine were tested with amikacin, amoxicillin-clavulanic acid, ampicillin, aztreonam, cefazolin, cefotaxime, ceftazidime, cefuroxime, ciprofloxacin, meropenem, netilmicin, piperacillin, and trimethoprim-sulfamethoxazole. *P. aeruginosa* and *Acinetobacter* sp. strains isolated from sources other than urine were tested with amikacin, aztreonam, ceftazidime, ciprofloxacin, meropenem, netilmicin, and piperacillin. Three thousand seven hundred twenty-four organism-antimicrobial agent combinations met the criteria and guidelines for susceptibility testing established by the NCCLS, and category interpretations were made according to those guidelines (8).

Disk diffusion susceptibility testing was performed according to the NCCLS guidelines (8). All inocula were prepared from the growth of pure cultures of bacteria cultivated for approximately 18 to 24 h on MacConkey agar II (Becton Dickinson Bioscience) or Columbia agar with 5% sheep blood (Becton Dickinson Bioscience) at 35°C. Bacterial isolates were suspended in 5 ml of brain heart infusion broth so that the turbidity was equivalent to that of a 0.5 McFarland

TABLE 1. Organism groups, organisms, and number of isolates tested

Organism group	Organism	No. of isolates
Escherichia coli	*Escherichia coli*	101
Klebsiella species	*Klebsiella oxytoca*	14
	Klebsiella pneumoniae	28
Proteus species	*Proteus mirabilis*	28
	Proteus vulgaris	2
Enterobacteriaceae with inducible β-lactamases	*Citrobacter freundii*	4
	Enterobacter species	37
	Morganella morganii	7
	Serratia liquefaciens	5
	Serratia marcescens	11
Pseudomonas aeruginosa	*Pseudomonas aeruginosa*	67
Acinetobacter species	*Acinetobacter* species	11

standard. All organisms were tested on one square Mueller Hinton II agar plate (Becton Dickinson Bioscience), as advised by Acar et al. (1). Antibiotics were applied to the plates by using antibiotic dispensers. These susceptibility testing plates were incubated in ambient air at 35°C. Inhibition zones were measured after 18 to 24 h of incubation by one person only. For each susceptibility test, visual ruler and video readings by both systems were made within a time period of 2 h. For both semiautomated systems, the procedure followed was that outlined in the instructions of each manufacturer. Because both manufacturers recommend a review of the results, zone diameters automatically measured by the video systems were, if necessary, adjusted on screen by the reader (reviewed readings). The visual reading obtained with a ruler was considered the "gold standard." The zone diameters calculated with the Osiris and Sirscan systems were compared to the zone diameters measured with a ruler according to the NCCLS standard method (8). Differences in zone diameter measurements (in millimeters) were recorded. In order to be able to compare our evaluation to the studies of Acar et al. (1) and Haddad-Prots et al. (3), we adopted the arbitrary threshold of 3 mm. That is, consensus was achieved when the results of the various systems did not differ by more than 3 mm. Interpretative categories (susceptible, intermediate, and resistant) were calculated for each zone measurement for each organism-antimicrobial agent combination tested. Discrepancies in interpretative categories were noted. A very major error was defined as a ruler reading interpretation of resistant and a video reading interpretation of susceptible. A major error was defined as a ruler reading interpretation of susceptible and a video reading interpretation of resistant. The following discrepancies were defined as minor errors: a ruler reading interpretation of resistant and a video reading interpretation of intermediate, a ruler reading interpretation of intermediate and a video reading interpretation of susceptible, a ruler reading interpretation of intermediate and a video reading interpretation of resistant, and a ruler reading interpretation of susceptible and a video reading interpretation of intermediate.

RESULTS

In total, 3,724 organism-antimicrobial agent combinations were evaluated prospectively.

For the interpretation, we divided the 315 organisms into six groups (Table 1): *E. coli* (101 strains), *Klebsiella* species (42 strains), members of the family *Enterobacteriaceae* with an inducible β-lactamase (64 strains), *Proteus* species (30 strains), *P. aeruginosa* (67 strains), and *Acinetobacter* species (11 strains).

The zone diameters were normally distributed for all organism-antimicrobial agent combinations. Because the visual ruler reading was considered the gold standard, the measure of agreement was determined with a population correlation coefficient in a constrained bivariate model (6). There were very good and similar correlations for the zone diameters of the reviewed readings for both the Osiris and the Sirscan systems versus the visual ruler readings. The analysis of variance for the Osiris and Sirscan systems resulted in intercepts of 0.51 and 0.51, respectively; slopes of 1.02 and 1, respectively; and correlation coefficients of 0.96 and 0.96, respectively. For all organism groups and antibiotics tested, the correlation coefficient exceeded 0.88 (Table 2) except for Osiris system readings with fosfomycin and Osiris and Sirscan system readings with amikacin.

Differences in zone measurements (in millimeters) between the video readings and the visual ruler readings were determined. Two thousand five hundred thirty-three (68.01%) reviewed readings for the Osiris system and 2,533 (68.01%) reviewed readings for the Sirscan system differed from the visual ruler readings. The mean sizes of these differences for the Osiris and Sirscan system readings versus the visual ruler readings were 2.31 ± 1.79 mm (standard deviation) and 2.19 ± 1.68 mm, respectively, with zone diameters systematically being slightly larger for the reviewed video readings. Differences that exceeded 3 mm were noted for 340 (9.12%) and 319 (8.56%) of the organism-antimicrobial agent combinations for the Osiris and Sirscan systems, respectively.

Interpretative category discrepancies were classified as very major errors, major errors, or minor errors (Table 3). The overall agreements for susceptibility interpretation were 96.82 and 94.59% for the Osiris and Sirscan systems, respectively, with simple kappa coefficients of 0.92 and 0.91 for the two systems, respectively.

Eight (1.07%) and 10 (1.34%) organism-antimicrobial agent combinations tested were found to be resistant by ruler reading and susceptible by video reading with the Osiris and the Sirscan systems, respectively, and were classified as very major errors. Two (0.07%) and 6 (0.21%) organism-antimicrobial agent combinations tested were found to be susceptible by ruler reading and resistant by video reading with the Osiris and the Sirscan systems, respectively, and were classified as major errors. The results for 118 and 124 organism-antimicrobial agent combinations tested were classified as minor errors with the Osiris and the Sirscan systems, respectively.

DISCUSSION

The Osiris and Sirscan semiautomated data management systems read, interpret, and report the antimicrobial agent disk diffusion susceptibility results.

The correlation of the zone diameter measurements of the reviewed Osiris system readings and the reviewed Sirscan system readings versus the visual ruler readings was good. The mean difference in the results obtained with both semiautomated systems in comparison with the results obtained by visual ruler reading was less than 2.5 mm. Zone diameters were systematically slightly larger for the video reading systems. In order to be able to compare our evaluation to the studies of Acar et al. (1) and Haddad-Prots et al. (3), we adopted the arbitrary threshold of a 3-mm zone diameter difference. In this setting, the results for 9.12 and 8.56% of the organism-antimicrobial agent combinations tested appeared to be discordant with the Osiris and Sirscan systems, respectively. These results

TABLE 2. Correlation of visual ruler readings versus reviewed video readings

Factor correlated	Osiris system			Sirscan system		
	Intercept	Slope	R^2	Intercept	Slope	R^2
Total correlation	0.51	1.02	0.96	0.51	1.00	0.96
Organism groups						
Escherichia coli	0.18	1.02	0.93	0.73	0.99	0.92
Klebsiella spp.	0.87	1.02	0.94	1.57	0.96	0.93
Enterobacteriaceae	0.24	1.01	0.98	0.19	1.01	0.97
Proteus spp.	1.81	0.97	0.94	1.18	0.99	0.94
Pseudomonas aeruginosa	0.23	1.07	0.94	0.20	1.03	0.96
Acinetobacter spp.	0.67	1.02	0.95	0.61	1.00	0.95
Drugs						
Ampicillin	0.15	1.01	0.96	−0.39	1.08	0.96
Amoxicillin-clavulanic acid	0.29	1.02	0.97	−0.03	1.00	0.98
Amikacin	2.02	0.94	0.85	3.23	0.89	0.80
Aztreonam	1.82	0.98	0.92	1.41	0.98	0.92
Cefazoline	1.07	0.93	0.89	0.94	0.93	0.90
Cefuroxime	0.18	1.02	0.96	0.04	1.00	0.98
Cefotaxime	0.69	1.01	0.96	0.39	0.99	0.96
Ceftazidime	0.55	1.01	0.92	1.60	0.96	0.89
Ciprofloxacin	0.32	1.07	0.95	0.01	1.04	0.96
Fosfomycin	3.29	0.90	0.82	3.90	0.87	0.88
Meropenem	0.25	1.04	0.95	1.02	0.98	0.94
Netilmicin	0.86	1.04	0.92	0.69	1.02	0.90
Nitrofurantoin	2.67	0.88	0.92	1.40	0.97	0.93
Norfloxacin	1.69	0.97	0.92	1.67	0.98	0.89
Piperacillin	1.27	0.97	0.95	1.44	0.98	0.94
Trimethoprim-sulfamethoxazole	0.76	1.04	0.93	0.91	1.01	0.93

are comparable to those reported by Acar et al. (1) and Haddad-Prots et al. (3).

In addition, these differences rarely affected the classification of the organisms as susceptible or resistant. The overall frequency of very major errors was low, i.e., 1.07% (8 of 742 combinations) and 1.34% (10 of 742 combinations) for the Osiris and Sirscan systems, respectively.

The study of Haddad-Prots et al. (3) compared Osiris system readings to manual caliper readings, and the studies of Medeiros et al. (7) and by Acar et al. (1) compared Sirscan system readings to manual (caliper) readings. The rates of very major errors reported in those three studies were comparable to those reported here (0.7, 0.3, and 1.76%, respectively). The differences between these studies are possibly due to the mix of species and antibiotics tested. Moreover, except for the *Acinetobacter* species strains, the discrepancies in results between bacterial species appeared to be random. A possible explanation for the discrepancies seen with the *Acinetobacter* species is the difficulty that the video systems have in visualizing the faint growth that sometimes occurs with this species. Another possible reason could be failure to swab the susceptibility plate thoroughly. If the growth of the organism is not confluent, the Osiris and Sirscan systems may read between the growth. An experienced technician can minimize these errors by reviewing the automatic readings prior to validation and adjusting them as needed. In this study the most likely reason for the discrepancies with the *Acinetobacter* species was the limited number of *Acinetobacter*-antibiotic combinations tested. More extensive evaluations with *Acinetobacter* species are recommended.

The antibiotics most often responsible for discrepant results were aztreonam, ciprofloxacin, meropenem, and norfloxacin. The inhibition zones obtained with aztreonam and ciprofloxa-

TABLE 3. Interpretative category discrepancies for visual ruler readings versus reviewed video readings

Discrepancy category	Proportion of false readings or errors[a]	
	Osiris system	Sirscan system
False readings by video system		
Ruler reading resistant vs video reading susceptible	8/742 (1.07)	10/742 (1.34)
Ruler reading susceptible vs video reading resistant	2/2,746 (0.07)	6/2,746 (0.21)
Minor errors by video system		
Ruler reading resistant vs video reading intermediate	21/742 (2.83)	35/742 (4.71)
Ruler reading intermediate vs video reading susceptible	55/236 (23.3)	50/236 (21.1)
Ruler reading intermediate vs video reading resistant	25/236 (10.5)	16/236 (6.77)
Ruler reading susceptible vs video reading intermediate	17/2,746 (0.61)	23/2,746 (0.83)

[a] A total of 315 organisms and 3,724 organism-antimicrobial agent combinations were tested with each system. The data represent the number of organism-antimicrobial agent combinations with a false reading or error/respective number of ruler readings (percent).

cin were large, which may be a possible source of the discrepant readings. The positions of the meropenem and norfloxacin disks in the corner of the petri dish also hamper precise measurement. These reasons for discrepant results for antibiotics with large inhibition zones and because of the position of the disk on the plate were also previously suggested by Haddad-Prots et al. (3).

In conclusion, the performances of the Osiris and Sirscan image analyzers are very similar and satisfactory. We think that both systems will be of great value in clinical practice.

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REFERENCES

1. **Acar, J., O. Bellon, H. Chardon, J. Charrel, F. Goldstein, R. Labia, and J. Sirot.** 1997. Statistical study of the Sirscan computerized camera. Pathol. Biol. **45:**383–388. (In French.)
2. **Berke, I., and P. M. Tierno, Jr.** 1996. Comparison of efficacy and cost-effectiveness of BIOMEC video and Vitek antimicrobial susceptibility test systems for use in the clinical microbiology laboratory. J. Clin. Microbiol. **34:**1980–1984.
3. **Haddad-Prots, L., D. Assayag, V. Jarlier, and J. Nguyen.** 1999. Evaluation of the Osiris video reader system for interpretation of disk diffusion susceptibility tests. Clin. Microbiol. Infect. **5**(Suppl. 3)**:**92.
4. **Jorgensen, J. H.** 1993. Selection criteria for an antimicrobial susceptibility testing system. J. Clin. Microbiol. **31:**2841–2844.
5. **Korgenski, E. K., and J. A. Daly.** 1998. Evaluation of the BIOMEC video reader system for determining interpretative categories of isolates on the basis of disk diffusion susceptibility results. J. Clin. Microbiol. **36:**302–304.
6. **Laurent, R. T.** 1998. Evaluating agreement with a gold standard in method comparison studies. Biometrics **54:**537–545.
7. **Medeiros, A. A., and J. Crellin.** 2000. Evaluation of the Sirscan automated zone reader in a clinical microbiology laboratory. J. Clin. Microbiol. **38:**1688–1693.
8. **National Committee for Clinical Laboratory Standards.** 1999. Performance standards for antimicrobial disk susceptibility testing. NCCLS document M2-A6, M100-S9. National Committee for Clinical Laboratory Standards, Wayne, Pa.