Accuracy of the BIOMIC Image Analysis Interpretation of Disk Diffusion Susceptibility Tests on Morphotypes of Pseudomonas aeruginosa From Cystic Fibrosis Patients

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Abstract

Background: Disk diffusion is the recommended method for susceptibility testing of P. aeruginosa from cystic fibrosis (CF) patients. The BIOMIC Image Analyzer (Giles Scientific, Santa Barbara, CA) for reading and interpretation of disk diffusion tests is a time and labor saving method over other susceptibility test methods. We compared the interpretation of disk diffusion results using the BIOMIC Image Analyzer with manual measurement of zone size diameters to determine if this device is a reliable method for interpreting the antibiotic susceptibilities of these bacteria and if it is affected by several inherent advantages.

Methods: One-hundred thirty-five isolates of P. aeruginosa were evaluated. Disk diffusion results were compared to a visual laboratory technician’s interpretation of zone size diameters. Agreement is calculated as a percentage of the totals as follows: SS, II, and RR are full agreement. Minor error (ME) is defined as erroneous categorization of a susceptible organism as resistant, and is calculated as SS/RR. Minor error (ME) is erroneous categorization of the true result (S or R) as intermediate and vice versa. Minor error (ME) is calculated as SS/RR. All proportions are presented as percentages and rounded to 1 decimal point. Acceptable agreement of a new method with a reference method is as follows:

1. Overall agreement >90%
2. Low very major error rate based on number of resistant isolates
3. The BIOMIC method provides great agreement with standard methods of reading disk diffusion plates by hand that are within FDA requirements. Test results are downloadable, providing protection from transcription errors, and greater speed of entry. Technicians involved with testing found it a simple, user-friendly method particularly for reading large numbers of tests.

Introduction

Disk diffusion is a long used, economical, and well validated method of determining bacterial susceptibility to several antibiotics at one time. Using a standardized laboratory technique it allows rapid and repeatable susceptibility testing.

The BIOMIC Image Analyzer is a commercially available automatic disk diffusion susceptibility reader. Software includes internal controls for consistency based on CLSI (previously NCCLS) guidelines, provides downloadable results, and flags units results for further analysis. The analyzer has a history of a great agreement with visual laboratory technician readings. The current study examined technician and machine agreement in reading disk diffusion susceptibility of P. aeruginosa strains isolated from CF patients.

Methods and Materials

Pseudomonas aeruginosa strains isolated from cystic fibrosis patients were tested against 12 antibiotics: aztreonam, ciprofloxacin, ceftazidime, cefepime, imipenem, meropenem, piperacillin/tazobactam, ticarcillin/clavulanate, ticarillin, and trimethoprim/sulfamethoxazole. Isolates were tested using disk diffusion methodology against 12 antibiotics: aztreonam, ciprofloxacin, ceftazidime, cefepime, imipenem, meropenem, piperacillin/tazobactam, ticarcillin/clavulanate, ticarillin, and trimethoprim/sulfamethoxazole. Isolates were inoculated into brain-heart infusion broth and incubated in ambient air at 37°C and diluted, as needed, to a 0.5 MacFarland standard. The bacterial suspension was inoculated onto a 150 mm Master agar plate by CLSI (recomend standard methodology) and the antibiotics applied over the surface. Plates were inoculated at 37°C in an incubator at 36 to 38°C. Zones of inhibition were measured by hand using callipers. The same plate was also read in the BIOMIC machine. Data were analyzed for agreements of new method with reference method results; 64% of the minor errors occurred with chloramphenicol and ciprofloxacin. Overall agreement was 97%. Chloramphenicol (11) and ciprofloxacin (7) were more frequently associated with major or very major errors.

Results

A total of 1620 antibiotic/organism combinations were tested from the three typical CF associated morphotypes: 36 classic, 53 mucoid, and 46 rough. There were 1,572 concordant results (97%) and 48 (<3%) discrepant results. Discrepant results were 28 (1.7%) minor errors, 8 (0.5%) major errors, and 12 (0.7%) very major errors. These error rates were well within error rates allowed by the FDA for approval of new systems.

Discussion

The BIOMIC method provides great agreement with standard methods of reading disk diffusion plates by hand that are within FDA requirements. Test results are downloadable, providing protection from transcription errors, and greater speed of entry. Technicians involved with testing found it a simple, user-friendly method particularly for reading large numbers of tests.