Comparison of BD Phoenix Automated Microbiology System with the MicroScan Rapid Neg ID Plus Neg MIC Panel Type 30 for Identification and Susceptibility Testing of Gram-negative Bacilli

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ABSTRACT

BACKGROUND: This study compared the new BD Phoenix (BD) Automated System with the widely used MicroScan Walkaway (MS) system for the identification and susceptibility testing of gram-negative bacilli.

METHODS: In the BD system NMIC/ID-26 panels were used. In the MS system Rapid Neg ID Panel Type 3 were used to identify the bacteria and Neg MIC Panel Type 30, with the turbidity method to prepare inoculums, were used for susceptibility testing. Both the BD and MS systems provided identification of the bacteria in < 4 hrs and susceptibility results after overnight incubation. When identification results were discrepant the correct identification was determined using API strips and tube biochemical tests. Discrepant susceptibility results, S in one system and R in the other, were resolved using microdilution MIC trays and standard NCCLS methods. A total of 262 isolates were tested.

RESULTS: Both systems identified 248 (95%) of the isolates to the same genus/species. The MS system required additional tests to identify 24 (9%) of these isolates. The MS system misidentified a Shigella species as a Providencia species and four isolates to different species within the same genera. The BD system misidentified an E. coli as S. flexneri, a Shigella species as E. coli, one E. cloacae isolate as C. brunii and one as K. ascorbata, and six isolates were identified as other species within the same genera. Susceptibility data were evaluated for 3,321 potentially clinically useful isolate-antimicrobial combinations. Both systems gave comparable results in 3,116 (94%). In 19 (0.57%), one system gave an S result and the other an R result. The reference system confirmed the BD result in 13 (68%) of these and the MS result in 6 (32%). There were 162 (4.9%) of the total where one system gave an S result and the other an I result, or an I/R combination occurred. In 119 (73%), the lower MIC occurred with the MS system.

CONCLUSION: Both the BD and MS Automated Systems provide efficient and effective methods for identification and susceptibility testing of gram-negative bacilli. When major susceptibility discrepancies occurred the reference method more often confirmed the BD result.

BACKGROUND

Timely identification and susceptibility testing of bacteria from patients’ specimens can have a significant impact on the management of infections. Many laboratories now use automated systems for testing bacteria in an efficient and cost-effective manner.

This study compared the new Becton Dickinson Phoenix (BD) Automated System with the widely used MicroScan Walkaway 96 S/I (MS) system or another rapid identification (ID) method for gram-negative bacilli. The study also compared the BD Phoenix with the MicroScan Walkaway 96 S/I system for antimicrobial susceptibility testing (AST).
TEST ISOLATES: A total of 262 isolates were tested, including Enterobacteriaceae, *Aeromonas* species, *Vibrio* species, and three commonly isolated species of glucose-nonfermenting gram-negative rods; 205 were current isolates from clinical specimens. Multiple isolates of the same species from the same patient were excluded. The 57 isolates from the laboratory’s frozen culture collection were selected so there was a representative sample of isolates for each genus/species to be tested. Isolates from the frozen collection were subcultured twice on sheep blood agar and incubated for 18 to 24 hours in ambient air at 35°C prior to testing. Inoculum suspensions for the BD Phoenix system and MicroScan system were set up on the same day using the same fresh 18-24 hour subculture plate. Separate purity plates were inoculated from both system’s basic inoculum. These subcultures were used to check purity and initiate discrepancy resolution.

THE BD PHOENIX SYSTEM. The BD Phoenix System consists of an instrument, software, disposable panels, broths for ID and AST, and an AST indicator. The ID method employs modified conventional, fluorogenic, and chromogenic substrates. The AST method is a broth based microdilution test that utilizes a redox indicator to enhance the detection of organism growth. The NMIC/ID-26 panels were used in this study. A 0.5 McFarland suspension of the test organism was made in the ID broth. The CrystalSpec Nephelometer was used to verify the density of the suspension and 25µL of this suspension was added to the AST broth. One drop of AST indicator was previously added to the AST broth. The suspension in the ID broth was used to inoculate the ID wells of the panel and the suspension in the AST broth was used to inoculate the AST wells. After loading the panels into the instrument, the panels are read at 20-minute intervals by the instrument. IDs, minimal inhibitory concentrations (MICs), and category interpretations are generated. Final results are available in 2-12 hours for ID and 4-16 hours for AST, however, the majority of IDs were completed in 2-3 hours and MICs in 6-8 hours. The Phoenix system also includes the BDXpert system software which analyzes ID and AST results against pre-defined rules and notifies the user of atypical results and patient conditions that may require further action.

THE MICROSCAN WALKAWAY 96 S/I SYSTEM. The MicroScan Walkaway 96 S/I system for identification and susceptibility testing includes an instrument for panel incubation and computerized result interpretation. The Rapid Neg ID Type 3 panel was used for ID. This is based on hydrolysis of fluorogenic substrates, pH changes following substrate utilization, and production of specific metabolic byproducts, or the rate of production of specific metabolic byproducts after 2.5 hours incubation in the instrument. A 0.5 McFarland suspension of the test organism was prepared in 6.5 mL of 0.4% saline with PLUORONIC® for inoculation of the ID panel. The Neg MIC Panel Type 30, with the turbidity standard technique for inoculum preparation, was used for AST. MICs are determined by the instrument after incubation of the panel for a minimum of 16 hours. A 0.5 McFarland suspension of the test organism was prepared in 3 mL of Inoculum Water, 100 µL of this suspension was pipetted into 25 mL of Inoculum Water with PLUORONIC®, and the resulting suspension was used for inoculation of the MIC panel. For both panel types, turbidity was confirmed with the MicroScan Turbidity Meter and rehydration and inoculation of the panels was performed using the MicroScan RENOX® Rehydrator/Inoculator system. The MicroScan system also includes the MicroScan LabPro Alert system software, which like the BD Expert system software, analyzes ID and AST results against pre-defined rules.

RAPID IDENTIFICATION METHODS. Rapid identification methods, as outlined in the National Committee for Clinical Laboratory Standards (NCCLS) M35-A2 Abbreviated Identification of Bacteria and Yeast document, were used for some isolates of *Escherichia coli*, swarming *Proteus* species, and *Pseudomonas aeruginosa* from non-sterile sites.

QUALITY CONTROL. The following quality control (QC) organisms were tested daily with the BD Phoenix system: *E. coli* ATCC 25922, *P. aeruginosa* ATCC 27853, *E. coli* ATCC 35218, and *Klebsiella pneumoniae* spp. *pneumoniae* ATCC 700603. Each new lot or shipment of MicroScan Rapid Neg ID Panel Type 3 was tested with *Klebsiella oxytoca* ATCC 49131, *Acinetobacter baumanii* ATCC 49139, *Shewanella putrefaciens* ATCC 49138, and *E. coli* ATCC 25922. MicroScan Neg MIC Panel Type 30 was tested weekly and with each new lot or shipment of panels with *E. coli* ATCC 25922, *P. aeruginosa* ATCC 27853, and *E. coli* ATCC 35218. Weekly QC of MicroScan MIC panels was instituted after QC was done for 30 consecutive test days.

DISCREPANCY TESTING OF ORGANISM IDENTIFICATIONS. The identification results from the BD Phoenix and MicroScan systems were compared for genus and species agreement. When the two systems agreed to the genus and species level, it was considered a correct identification. When the species were not in agreement for the two systems, the isolate was retested (in duplicate) in parallel in both systems. When discrepancy remained, the organism was tested with API 20E strips (bioMerieux, Inc. Durham, NC) and/or conventional tube biochemicals (arbitration method). The identification (BD Phoenix or MicroScan) that agreed with the result of the arbitration method was considered the correct identification.

DISCREPANCY TESTING OF SUSCEPTIBILITIES. Only those antibiotics that were tested in both systems were considered and category concordance evaluated. When a meaningful error existed, the isolate was retested for the antibiotic(s) in question in parallel in both systems; duplicate tests were done. When the results remained discrepant, the isolate was tested with microdilution trays prepared according to procedures of the NCCLS and frozen at -70°C until use. Each inoculum was prepared by growing the organism in brain heart infusion broth for several hours, diluting the inoculum in 0.2% Tween 80 in sterile water, and inoculating the trays with a MIC 2000 automatic inoculator (Thermo Labsystems, MA) to yield a final concentration of approximately 5 x 10^5 CFU/mL. The MICs were determined, after 16–24 hours incubation, by observing the presence or absence of visible growth under transmitted light.
Organism identification. Both systems identified 248 (95%) of the isolates to the same genus/species (Table 1). The MS system misidentified a Shigella species as a Providencia species and four isolates to different species within the same genera. The BD system misidentified an E. coli as S. flexneri, a Shigella species as E. coli, one Enterobacter cloacae isolate as Citrobacter braakii and one as K. oxytoca, and six isolates were identified as other species within the same genera (Table 2).

The MicroScan system required additional tests to identify 24 (9%) of these isolates and did not separate the following species of organisms: Acinetobacter baumannii, Acinetobacter lwoffii, Acinetobacter lwoffii, Aeromonas hydrophila, and Citrobacter braakii. The MS system did not require additional tests for identification of any of the isolates.

Antimicrobial susceptibility testing. BD Phoenix and MicroScan discrepancies for AST were categorized as follows (Table 3):

- Categorical agreement and + 1 dilution difference
- Intermediate (I) in one system, Susceptible (S) or Resistant (R) in the other
- Meaningful errors—one system Susceptible and the other Resistant
- Comparable results—categorical agreement and + 1 dilution agreement

Susceptibility data were evaluated for 3,321 potentially clinically useful isolate-antimicrobial combinations. Both systems gave comparable results in 3,116 (94%). In 19 (0.57%), one system gave a Susceptible result and other a Resistant result. The reference system confirmed the BD Phoenix result in 13 (68%) of these and the MicroScan result in 6 (32%). There were 162 (4.9%) of the total where one system gave an Intermediate result and the other a Susceptible or Resistant result. Both systems’ panels screen for extended spectrum beta lactamase (ESBL) production. Thirty four E. coli isolates yielded negative results on both systems. Of the 26 K. oxytoca and K. pneumoniae isolates tested, 24 yielded negative results on both systems and 1 yielded a positive result on both systems. One isolate yielded a negative result on the MicroScan and a possible ESBL result on the BD Phoenix. This isolate yielded a negative result when tested by the NCCLS disk method for detection of ESBL production. Due to only one isolate yielding a positive result, further testing is required to evaluate the BD Phoenix system’s ability to detect ESBL production.
CONCLUSIONS

- Both the BD Phoenix and MicroScan automated systems provide efficient and effective methods for the identification and susceptibility testing of gram-negative bacilli.

- When major susceptibility discrepancies occurred, the reference method more often confirmed the BD Phoenix result.

- The MicroScan system is more likely to require supplemental biochemical tests to identify the bacteria.