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ARTICLE

E. Stefaniuk · A. Baraniak · M. Gniadkowski · W. Hryniewicz

Evaluation of the BD Phoenix Automated Identification and Susceptibility Testing System in Clinical Microbiology Laboratory Practice

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Abstract The Phoenix Automated Microbiology System (BD Biosciences, USA) is a new, fully automated system for the rapid identification and antimicrobial susceptibility testing of gram-positive and gram-negative bacteria. The objective of this study was to evaluate the quality of performance of the Phoenix system in the identification and antimicrobial susceptibility testing of 260 gram-negative (n=174) and gram-positive (n=86) isolates collected from Polish hospitals in recent years. Two Phoenix panel types for identification/antimicrobial susceptibility testing, NMIC/ID-5 for gram-negative rods and PMIC/ID-4 for gram-positive cocci, were used in the analysis according to the manufacturer's recommendations. The results produced by the system were compared with data obtained by reference or conventional microbiological methods. A high rate of agreement between the Phoenix and the conventional methods was observed for identification, ranging from 100% for gram-positive cocci to 96.0% for gram-negative nonfermenters and 92.5% for members of the family *Enterobacteriaceae*. Similarly, a high level of agreement characterized the antimicrobial susceptibility data obtained with the Phoenix and by the agar dilution method (2,361 test results). For staphylococci, enterococci and *Enterobacteriaceae*, the methods were 100% concordant in determining the category of susceptibility of isolates to the majority of the antimicrobial agents tested. A category agreement value of below 90% was found for the susceptibility of enterococci and gram-negative nonfermenters to ciprofloxacin (84.6% and 88.5%, respectively) and for susceptibility of *Stenotrophomonas maltophilia* to trimethoprim-sulfamethoxazole (80.0%).

E. Stefaniuk (✉) · W. Hryniewicz
Department of Epidemiology and Clinical Microbiology,
National Institute of Public Health,
Chelmska 30/34, 00-725 Warsaw, Poland
e-mail: estefaniuk@cls.edu.pl
Tel.: +48-22-8413367
Fax: +48-22-8412949

A. Baraniak · M. Gniadkowski
Department of Molecular Microbiology,
National Institute of Public Health,
Chelmska 30/34, 00–725 Warsaw, Poland

Introduction

Rapid identification (ID) and antimicrobial susceptibility testing (AST) of clinically significant bacterial isolates are required for the proper management of infected patients. This has become particularly important in recent decades, mainly due to the alarming spread of drug resistance in pathogenic bacteria. One way to shorten the time consumed by microbiological analyses is automation of the laboratory practice. The first semiautomated and fully automated ID/AST systems were introduced approximately 30 years ago. However, changes in the spectrum of etiological agents and in their susceptibility profiles demand the continuous improvement of such systems and their adjustment to new epidemiological situations.

The Phoenix Automated Microbiology System (BD Biosciences, USA) is a new, fully automated system for rapid ID and AST of gram-positive and gram-negative bacteria. It can analyze up to 100 ID and AST combined panels (ID/AST panels) simultaneously. The time needed for obtaining a complete set of the ID/AST results is short (8–12 h) and varies with the species being tested. In this study, we used the Phoenix to analyze groups of varied gram-negative and gram-positive isolates from Polish hospitals. The results of the analyses were compared with those obtained by conventional microbiological methods.

Materials and Methods

Bacterial Strains

A total of 260 clinically significant gram-negative (n=174) and gram-positive (n=86) bacterial pathogens were included in the study. Gram-negative organisms represented eight of the most commonly encountered species of the family *Enterobacteriaceae*

Table 1 Bacterial species and numbers of isolates of each particular species used in the study

Species	No. of isolates
Enterobacteriaceae	
Escherichia coli	32
Klebsiella pneumoniae	18
Klebsiella oxytoca	1
Enterobacter cloacae	27
Serratia marcescens	10
Citrobacter freundii	16
Morganella morganii	5
Proteus mirabilis	11
Gram-negative nonfermenting rods	
Pseudomonas aeruginosa	22
Acinetobacter baumannii	17
Stenotrophomonas maltophilia	15
Staphylococci	
Staphylococcus aureus	24
Staphylococcus epidermidis	7
Staphylococcus haemolyticus	4
Staphylococcus saprophyticus	2
Enterococci	
Enterococcus faecalis	21
Enterococcus faecium	18
Enterococcus casseliflavus	5
Enterococcus gallinarum	5

and three species of nonfermenting rods. Gram-positive isolates belonged to four *Staphylococcus* species and four species of the genus *Enterococcus*. The bacterial species and numbers of isolates of each particular species analyzed are shown in Table 1. The isolates were recovered from various specimens collected from patients hospitalized in different medical centers in Poland from 1997 to 2000. They were not epidemiologically related. A wide variety of clinically important antimicrobial resistance mechanisms were represented by the isolates, including extended-spectrum β-lactamases (ESBLs) in *Enterobacteriaceae* (48 isolates), methicillin resistance in staphylococci (12 isolates), and resistance to vancomycin and to high concentrations of aminoglycosides in enterococci (11 and 33 isolates, respectively).

Conventional Identification Methods

Gram-negative isolates were identified to the species level by the API 20 E (*Enterobacteriaceae*) or the API 20 NE (nonfermenters) test (bioMérieux, France). *Staphylococcus aureus* was identified by coagulase production (bound and free), and coagulase-negative staphylococci were speciated by the API Staph test (bioMérieux). Enterococci were identified to the genus level according to the method of Facklam and Collins [1] and to the species level by the API 20 Strep test (bioMérieux), supplemented by the potassium tellurite reduction, motility, and pigment production tests [1].

Conventional Antimicrobial Susceptibility Testing

Minimal inhibitory concentrations (MICs) of various antimicrobial agents were evaluated by the agar dilution method in accordance with the National Committee for Clinical Laboratory Standards (NCCLS) guidelines [2, 3]. The antimicrobial agents tested were the same as those that were present in the Phoenix panels used, and specific sets of agents were selected for each of the species analyzed. The following antimicrobial agents were included in the study: penicillin, ampicillin, cefotaxime, rifampin, streptomycin, gentamicin, and tetracycline (Polfa Tarchomin, Poland); amoxicillin, clavulanic acid, oxacillin and ceftazidime (Glaxo SmithKline, UK); piperacillin, and tazobactam (Wyeth, USA); cefepime and amikacin (Bristol-Myers Squibb, USA); meropenem (Zeneca, UK); clindamycin (Pharmacia & Upjohn, Belgium); ciprofloxacin (Bayer, Germany), tobramycin and vancomycin (Eli Lilly, USA); teicoplanin (Marion Merrell, UK); and trimethoprim-sulfamethoxazole (Roche, Switzerland). In β-lactam-inhibitor combinations, the concentrations of inhibitors used were in accordance with NCCLS guidelines [3]. *Escherichia coli* ATCC 25922, *Escherichia coli* ATCC 35218, *Pseudomonas aeruginosa* ATCC 27853, *Staphylococcus aureus* ATCC 29213, and *Enterococcus faecalis* ATCC 29212 were used as reference strains.

Detection of Methicillin Resistance in Staphylococci

Methicillin-resistant staphylococci were detected by two methods recommended by the NCCLS: the 1 μg oxacillin disk and the agar screening method [3, 4]. The screening method was used only for *Staphylococcus aureus* isolates. Results of the analysis were confirmed by polymerase chain reaction (PCR) amplification of the *mecA* gene in all the methicillin-resistant isolates as described previously [5]. *Staphylococcus aureus* ATCC 25923, *Staphylococcus aureus* ATCC 43300, and *Staphylococcus aureus* MR3 from the collection of the National Institute of Public Health in Warsaw were used as reference strains.

Detection of High-Level Aminoglycoside Resistance in Enterococci

Enterococcal isolates were tested for the presence of the high-level aminoglycoside resistance (HLAR) phenotype by the agar screening method as described by the NCCLS [3, 4]. *Enterococcus faecalis* ATCC 29212 and *Enterococcus faecalis* ATCC 51299 were used as reference strains.

Detection of Vancomycin Resistance in Enterococci

Vancomycin-resistant enterococci were identified by the NCCLS agar screening procedure [2, 4], and positive results of the test were confirmed by PCR detection of *vanA* or *vanB* genes as described previously [6, 7]. *Enterococcus faecalis* ATCC 29212 and *Enterococcus faecalis* ATCC 51299 were used as reference strains.

Detection of Extended-Spectrum β-Lactamases in Enterobacteriaceae

ESBLs were detected in *Enterobacteriaceae* isolates by the double-disk synergy (DDS) test [8] with disks containing cefotaxime, ceftazidime, and amoxicillin-clavulanate. The disks were placed 20 mm apart (from center to center). The 48 DDS-positive isolates included 15 *Klebsiella pneumoniae*, 15 *Escherichia coli*, 10 *Serratia marcescens*, 6 *Enterobacter cloacae*, and 2 *Citrobacter freundii* isolates. They were recovered in seven Polish hospitals from 1996 to 1998 and were confirmed to be ESBL producers by biochemical and molecular methods (isoelectric focusing, bioassay, PCR, and sequencing) in previous works [9, 10]. *Escherichia coli* ATCC 25922, *Escherichia coli* ATCC 35218, and *Klebsiella pneumoniae* ATCC 700603 were used as reference strains.

Identification and Antimicrobial Susceptibility Testing by Phoenix

Two Phoenix ID/AST panel types, NMIC/ID-5 for gram-negative rods (European Gram-Negative Combo, ref. no. 448510) and PMIC/ID-4 for gram-positive cocci (European Gram-Positive Combo, ref. no. 448508), were used in the analysis according to the manufacturer's recommendations. Bacterial suspensions were prepared from overnight cultures grown on Columbia agar plates

Table 2 Agreement of the identification (ID) data obtained with the Phoenix and with standard methods

Species (no. of isolates)	ID agreement between standard methods and Phoenix (no. of isolates)	Discrepant ID at genus level (no. of isolates)	Discrepant ID at species level (no. of isolates)	ID by Phoenix in discrepant cases[a]
Proteus mirabilis (11)	9	2	0	*Pasteurella multocida* *Alcaligenes faecalis*
Enterobacter cloacae (27)	25	0	2	*Enterobacter sakazakii* *Enterobacter gergoviae*
Citrobacter freundii (16)	14	0	2	*Citrobacter farmerii* *Citrobacter braaki*
Serratia marcescens (10)	7	1	2	*Serratia plymutica* (*n*=2) *Pantoea agglomerans*
Pseudomonas aeruginosa (22)	20	0	2	*Pseudomonas* spp. *Pseudomonas putida*

[a] Only the discrepant cases are presented here. Complete agreement of the identification at both the genus and the species level was observed for *Staphylococcus aureus*, *Staphylococcus epidermidis*, *Staphylococcus haemolyticus*, *Staphylococcus saprophyticus*, *Enterococcus faecalis*, *Enterococcus faecium*, *Enterococcus casseliflavus*, *Enterococcus gallinarum*, *Escherichia coli*, *Klebsiella pneumoniae*, *Klebsiella oxytoca*, *Morganella morganii*, *Acinetobacter baumannii* and *Stenotrophomonas maltophilia*

with 5% sheep blood (BD Biosciences, ref. no. 2504071) and adjusted to a density of 0.5–0.6 McFarland in 4.5 ml of Phoenix ID Broth using a BD CrystalSpec nefelometer (BD Biosciences). Phoenix AST Broth was supplemented with Phoenix AST Indicator (1 drop per tube) and used for dilution of the initial bacterial suspensions to the final density of 5×10^5 cfu/ml. This was achieved by adding 25 µl of the initial suspensions to 8.0 ml of the AST broth. The initial suspensions were then used for inoculation of the ID parts of ID/AST panels, whereas their AST parts were inoculated with the AST broth suspensions. The panel preparation time did not exceed 30 min, and inoculated panels were placed in the Phoenix instrument for incubation and automated reading of the results.

Evaluation of the Phoenix Identification and Antimicrobial Susceptibility Testing Results

The Phoenix ID and AST results were compared with those obtained by reference or conventional microbiological methods. The ID agreement was calculated individually for gram-positive cocci, *Enterobacteriaceae*, and gram-negative nonfermenting rods as the ratio of the number of isolates for which identical results were obtained to the number of all isolates tested within each of the groups. The lack of conformity in ID was considered separately at the genus and species levels.

The concordance of the two methods for classification of an isolate into the resistant (R), intermediate (I), or susceptible (S) category with regard to each antimicrobial agent used (according to NCCLS criteria [3]) was analyzed. The category agreement rates for particular agents were calculated separately for staphylococci, enterococci, *Enterobacteriaceae*, and gram-negative nonfermenters. Any classification discrepancy between Phoenix and the agar dilution method was interpreted either as a "major error" (ME) (R by Phoenix and S by the reference method), "very major error" (VME) (S by Phoenix and R by the reference method,) or "minor error" (mE) (I by one method and S or R by the other method). The number of these errors was determined for each antimicrobial agent for each of the groups of species tested. AST results were compared for 249 isolates for which complete ID agreement was obtained.

The ability of the Phoenix method to detect methicillin resistance in staphylococci, HLAR and vancomycin resistance in enterococci, and ESBLs in *Enterobacteriaceae* was evaluated in terms of sensitivity, specificity, and agreement with reference or conventional methods. Sensitivity was calculated as the ratio of the number of Phoenix true-positive results to the number of standard positive results, whereas specificity was defined as the ratio of the number of Phoenix true-negative results to the number of standard negative results. Agreement was calculated as the ratio of the number of Phoenix true-positive and true-negative results to the number of all tests performed.

Results

Identification of Isolates

The results of the comparison of the ID data obtained with the Phoenix and by conventional methods are shown in Table 2. A high rate of agreement between the Phoenix and the conventional methods was observed. It ranged from 100% for gram-positive cocci to 96.0% for gram-negative nonfermenters and 92.5% for *Enterobacteriaceae* isolates (Table 2). Considering all the isolates tested together, the overall rate of agreement was 95.8%. Discrepancies between the results obtained by the Phoenix and those obtained by conventional methods were observed in 11 cases. The majority of these discrepancies occurred at the species level. A discrepancy at the genus level was observed in three *Enterobacteriaceae* isolates, identified by the API 20 E as *Proteus mirabilis* (*n*=2 isolates) and *Serratia marcescens* but identified by the Phoenix as *Pasteurella multocida*, *Alcaligenes faecalis*, and *Pantoea agglomerans*, respectively. In each of the discrepant cases, the procedure was repeated three times by both methods, yet the same results were obtained. The Phoenix properly identified all *Enterococcus casseliflavus* and *Enterococcus gallinarum* isolates as *Enterococcus casseliflavus/Enterococcus gallinarum*, adding a comment that the additional test for yellow pigment production should be performed in order to discriminate between the two species.

Antimicrobial Susceptibility Testing

Results of the analysis, shown in Tables 3 and 4, revealed a high level of concordance between the Phoenix data and

Table 3 Category agreement (CA) of the antimicrobial susceptibility data obtained with the Phoenix and by reference methods: gram-positive isolates

Species and antimicrobial agent	No. of tests	CA [no. of tests (%)][a]	Type of discrepancy[b, c]		Errors		
			Phoenix	Reference method	mE (%)	ME (%)	VME (%)
Staphylococcus spp.							
Gentamicin	37	35 (94.6)	*S. epidermidis* R (MIC, 16) I (MIC, 8)	*S. epidermidis* I (MIC, 8) R (MIC, 16)	5.4	0	0
Ciprofloxacin	37	36 (97.3)	*S. epidermidis* S (MIC, $\leq$1)	*S. epidermidis* I (MIC, 2)	2.7	0	0
Enterococcus spp.							
Tetracycline	39	38 (97.4)	*E. faecium* S (MIC, 4)	*E. faecium* R (MIC, 16)	0	0	2.6
Ciprofloxacin	39	33 (84.6)	*E. faecium* I (MIC, 2) I (MIC, 2)	*E. faecium* R (MIC, 8) R (MIC, 4)	12.8	2.6	0
			E. faecalis I (MIC, 2) R (MIC, 4) S (MIC, $\leq$1) I (MIC, 2)	*E. faecalis* S (MIC, 1) S (MIC, 1) I (MIC, 2) S (MIC, 1)			

mE, minor error; ME, major error; VME, very major error

[a] Number of tests refers to the tests in which concordant results were obtained by the Phoenix and the agar dilution method

[b] Only the discrepant cases are shown here. Complete agreement of the susceptibility data was observed in the case of staphylococci and oxacillin, clindamycin, tetracycline, trimethoprim-sulfamethoxazole, rifampin, vancomycin and teicoplanin; and in the case of enterococci and penicillin, gentamicin, streptomycin, vancomycin, and teicoplanin

[c] This column presents (i) species of the isolates for which discrepancies between the Phoenix and the agar dilution method were observed, (ii) susceptibility categories for the respective species as indicated by each method, and (iii) the MICs of the antimicrobial agents (μg/ml)

those obtained by the agar dilution method. For staphylococci, enterococci, and *Enterobacteriaceae*, the 100% agreement of susceptibility categorization was observed with the majority of antimicrobial agents tested. A category agreement value of below 90% was found only in the case of ciprofloxacin susceptibility of enterococci (84.6%) and gram-negative nonfermenters (88.5%), and in the case of trimethoprim-sulfamethoxazole susceptibility of *Stenotrophomonas maltophilia* (80.0%). The majority of discrepancies between the two methods could be interpreted as minor errors. Major errors were obtained in the cases of ciprofloxacin susceptibility of single *Enterococcus faecalis* and *Pseudomonas aeruginosa* isolates, gentamicin susceptibility of a single *Pseudomonas aeruginosa* isolate, and trimethoprim-sulfamethoxazole susceptibility of two *Stenotrophomonas maltophilia* isolates. Very major errors occurred in the tetracycline susceptibility evaluation of a single *Enterococcus faecium* isolate, and in the trimethoprim-sulfamethoxazole susceptibility testing of single *Escherichia coli* and *Stenotrophomonas maltophilia* isolates.

Detection of Resistance Phenotypes

Table 5 shows results of the detection of methicillin resistance in staphylococci, of HLAR and vancomycin resistance in enterococci, and of ESBLs in *Enterobacteriaceae*. In the case of the three resistance phenotypes in gram-positive cocci, the Phoenix demonstrated 100% sensitivity, specificity, and agreement with the reference methods. The detection of ESBLs in enterobacteria was not as accurate, with the following values observed: sensitivity of 95.8%, specificity of 96.2%, and agreement of 96% when compared with the DDS test. The false-negative results were obtained with two *Enterobacter cloacae* isolates that produced the CTX-M-3 ESBL [11] and in which the expression of the AmpC cephalosporinase was inducible [12]. The DDS-negative and Phoenix-positive results were observed with two other *Enterobacter cloacae* isolates for which high MICs of cefotaxime and ceftazidime (32–64 μg/ml) and comparably high MICs of piperacillin and piperacillin plus tazobactam (128 and 64–128 μg/ml, respectively) were observed. This data suggested that the AmpC cephalosporinase derepression [13] mediated the phenotype; this indication was confirmed by β-lactamase isoelectric focusing and bioassay (results not shown).

Discussion

Very few reports evaluating the accuracy of the ID and AST analyses performed with the Phoenix system have been published to date. Brisse et al. [14] compared the Phoenix and the Vitek 2 (bioMérieux) in the ID of clinical isolates of the *Burkholderia cepacia* complex. They found the results obtained with both systems unsatisfactory and

Table 4 Category agreement (CA) of the antimicrobial susceptibility data obtained with the Phoenix and by reference methods: gram-negative isolates

Species and antimicrobial agent	No. of tests	CA [no. of tests (%)][a]	Type of discrepancy[b, c]		Errors		
			Phoenix	Reference method	mE (%)	ME (%)	VME (%)
Enterobacteriaceae							
Amoxicillin-clavulanic acid	65	63 (96.9)	*C. freundii* I (MIC, 16) (x2)	*C. freundii* R (MIC, 32) (x2)	3.1	0	0
Piperacillin-tazobactam	111	107 (96.4)	*K. pneumoniae* R (MIC, >64) *C. freundii* S (MIC, 16) R (MIC, >64) *E. coli* I (MIC, 64)	*K. pneumoniae* I (MIC, 64) *C. freundii* I (MIC, 32) I (MIC, 64) *E. coli* S (MIC, 16)	3.6	0	0
Cefotaxime	111	110 (99.1)	*E. coli* I (MIC, 16)	*E. coli* S (MIC, 4)	2.6	0	0
Ciprofloxacin	65	63 (96.9)	*E. cloacae* I (MIC, 2) I (MIC, 2)	*E. cloacae* R (MIC, >16) S (MIC, 0.12)	3.1	0	0
Trimethoprim-sulfamethoxazole	65	64 (98.5)	*E. coli* S (MIC, <0.5)	*E. coli* R (MIC, >64)	0	0	1.5
Gram-negative nonfermenting rods							
Cefotaxime	52	51 (98.1)	*S. maltophilia* I (MIC, 16)	*S. maltophilia* S (MIC, 8)	1.9	0	0
Ceftazidime	52	50 (96.2)	*P. aeruginosa* I (MIC, 16) S (MIC, 4)	*P. aeruginosa* S (MIC, 4) I (MIC, 16)	3.8	0	0
Cefepime	52	48 (92.3)	*S. maltophilia* R (MIC, >16) I (MIC, 16) *P. aeruginosa* R (MIC, >16) I (MIC, 16)	*S. maltophilia* I (MIC, 16) S (MIC, 8) *P. aeruginosa* I (MIC, 16) S (MIC, 8)	7.0	0	0
Amikacin	52	51 (98.1)	*S. maltophilia* S (MIC, 16)	*S. maltophilia* I (MIC, 32)	1.9	0	0
Gentamicin	52	48 (92.3)	*S. maltophilia* I (MIC, 8) *P. aeruginosa* R (MIC, >8) R (MIC, >8) I (MIC, 8)	*S. maltophilia* S (MIC, 4) *P. aeruginosa* S (MIC, 2) I (MIC, 8) S (MIC, 4)	5.8	1.9	0
Tobramycin	52	51 (98.1)	*S. maltophilia* I (MIC, 8)	*S. maltophilia* S (MIC, 4)	1.9	0	0
Ciprofloxacin	52	46 (88.5)	*S. maltophilia* I (MIC, 2) (x2) I (MIC, 2) I (MIC, 2) *P. aeruginosa* R (MIC, >2) S (MIC, $\leq$0.25)	*S. maltophilia* S (MIC, 1) R (MIC, 4) R (MIC, 8) *P. aeruginosa* S (MIC, 1) I (MIC, 2)	9.6	1.9	0
Trimethoprim-sulfamethoxazole[d]	15	12 (80.0)	*S. maltophilia* S (MIC, 2/38) R (MIC>4/76) (x2)	*S. maltophilia* R (MIC, 8/152) S (MIC, 2/38) (x2)	0	13.3	6.7

mE, minor error; ME, major error; VME, very major error

[a] Number of tests refers to the tests in which concordant results were obtained by Phoenix and the agar dilution method

[b] Only discrepant cases are shown here. Complete agreement of the susceptibility data was observed in the case of *Enterobacteriaceae* and ampicillin, piperacillin, ceftazidime, cefepime, meropenem, amikacin, gentamicin, and tobramycin; and in the case of gram-negative nonfermenting rods and piperacillin, piperacillin with tazobactam, and meropenem

[c] This column presents (i) species of the isolates for which discrepancies between the Phoenix and the agar dilution method were observed, (ii) susceptibility categories for the respective species as indicated by each method, and (iii) the MICs of the antimicrobial agents (μg/ml)

[d] Not tested by the agar dilution method for *Pseudomonas aeruginosa* and *Acinetobacter baumannii*

Table 5 Selected resistance phenotypes detected by the Phoenix and by standard methods

	Resistance phenotype			
	MRS	HLAR	VRE	ESBL
No. of isolates positive by standard methods and by the Phoenix	12	33	11	46
No. of isolates negative by standard methods and by the Phoenix	25	6	28	50
No. of isolates positive by standard methods and negative by the Phoenix	0	0	0	2
No. of isolates negative by standard methods and positive by the Phoenix	0	0	0	2
Agreement (%)	100	100	100	96.0
Sensitivity (%)	100	100	100	95.8
Specificity (%)	100	100	100	96.2

MRS, multiresistant staphylococci; HLAR, high-level aminoglycoside resistance; VRE, vancomycin-resistant enterococci; ESBL, extended-spectrum beta-lactamases

postulated molecular biology methods as the best tool for the differentiation of these organisms. Endimiani et al. [15] used the Phoenix in the ID and AST analysis of 136 isolates of various gram-negative nonfermenting rods. They compared the ID data with those obtained with the ATB system (bioMérieux), and the AST results were compared with those obtained by the broth microdilution method. A high rate of agreement in ID (97.1%) and a lack of major errors in AST were observed, which prompted the authors to describe the Phoenix as a reliable diagnostic system. Leverstein-van Hall et al. [16] compared the Phoenix, the Vitek 1 (bioMérieux), and the Vitek 2, using the E test (AB Biodisk, Sweden) as a reference method in the detection of ESBLs in 74 *Escherichia coli* and *Klebsiella* isolates recovered in a single hospital. The authors concluded that, with a sensitivity of 89% and a specificity of 87%, the Phoenix was the most accurate system in that particular study [16].

The analysis reported here was performed on a more diverse group of bacterial isolates than in the earlier studies. It included both gram-positive and gram-negative pathogens representing various susceptibility profiles. With the total ID agreement with API tests of 95.8% and only three very major errors in AST when compared with the reference method (out of a total of 2,361 test results), the Phoenix performed well in this study. This observation is additionally strengthened if the high rates of sensitivity, specificity, and agreement with the reference methods in the detection of several important resistance phenotypes (methicillin resistance in staphylococci, HLAR and vancomycin resistance in enterococci, and ESBLs in *Enterobacteriaceae*) are considered. The ID performance was especially good in the case of gram-positive cocci and gram-negative nonfermenters; the lowest rate of agreement with the API tests (92.5%) was observed with isolates of *Enterobacteriaceae*.

However, even with enterobacteria, only three of the nine discrepant cases (120 isolates altogether) occurred at the genus level. In the AST analysis, the performance of the Phoenix system with staphylococci (no major or very major errors) and *Enterobacteriaceae* (no major errors, 1 very major error) was excellent. A lower level of accuracy was observed with enterococci and gram-negative nonfermenting rods, for which both major and very major errors were obtained. These errors concerned specific antimicrobial agents that are rarely used for treatment of an emerging infection, i.e. tetracycline and ciprofloxacin in the case of enterococci, and gentamicin, ciprofloxacin, and, in particular, trimethoprim-sulfamethoxazole in the case of nonfermenters. It is difficult to explain the possible source of the repeated major and very major errors, especially those obtained with trimethoprim-sulfamethoxazole and single isolates of *Escherichia coli* and *Stenotrophomonas maltophilia*. With regard to the important resistance phenotypes, the only cases of discrepancy between the Phoenix and the reference methods were in the detection of ESBLs. The two false-negative and two false-positive results were obtained with isolates of *Enterobacter cloacae*, one of the species known to produce species-specific AmpC cephalosporinases [13]. These enzymes may complicate the detection of ESBLs, especially detection by quantitative procedures (used in automatic systems). However, it is impossible to establish why the Phoenix repeatedly produced false-positive results with the two AmpC-derepressed *Enterobacter cloacae* isolates, neither of which responded to β-lactamase inhibitors.

The work reported here was carried out on a highly diverse group of clinically important microorganisms. To our knowledge, this is the first evaluation of the Phoenix that includes gram-positive pathogens. Nevertheless, further studies in clinical microbiology laboratories are warranted to confirm these findings.

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