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Evaluation of the Sensititre Aris® 2X, Vitek® 2 and Phoenix® Automated Systems for Antimicrobial Susceptibility Testing of Gram-Positive Pathogens Originating from a Regional Healthcare Network

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REVISED ABSTRACT*

Background: Reliable detection of resistance is mandatory for patient management and infection control tracking. Given the widespread use of automated systems, continued performance assurance requires periodic re-evaluation. We evaluated the accuracy of the Sensititre ARIS 2X (TREK Diagnostic Systems) and Vitek 2 (bioMérieux) systems when testing Gram-positive cocci.

Methods: The challenge collection consisted of contemporary staphylococci (98), streptococci (12) and enterococci (52) recovered from skin and soft tissue (58%), urine (31%), blood (5.6%) and others (5.6%). Isolates were tested using the Sensititre susceptibility plate (Part No. GPALL1F) and Vitek 2 (Part No. GP67) assay panels, and were compared with the existing algorithm consisting of the Phoenix PMIC-106 panel, disk diffusion and ETest (for group B streptococci). Consensus was defined as all three systems with matching CLSI categorical results.

Results: A total of 3,897 results were generated with 13 antimicrobial agents. Overall agreement was 92.8%. Those agents displaying >=95% agreement included gentamicin (100), nitrofurantoin (96.0), tetracycline (96.3), trimethoprim/sulfamethoxazole (100) and vancomycin (99.3). Total error rates were 3.0%, 1.8% and 2.0% for the Phoenix, Sensititre ARIS 2X and Vitek 2, respectively. Error rates for those agents displaying <95% agreement are in the Table.

Conclusions: Acceptable categorical agreement (92.8% overall) was achieved with the antimicrobial test systems for isolates originating in 2010. Errors were more evident when testing certain species (staphylococci) and antimicrobial agents (oxacillin, cefoxitin screen, levofloxacin) with particular systems: Phoenix (3.0% total error) > Vitek 2 (2.0%) > Sensititre (1.8%).

			% Error Rate		
Antimicrobial Agent	**n**	**% Agreement**	**Phoenix**	**Sensititre ARIS 2X**	**Vitek 2**
Ampicillin	52	94.2	1.9	0.0	3.8
Cefoxitin Screen	90	77.8	7.8	13.3	1.1
Clindamycin	101	91.1	1.0	3.0	2.0
Erythromycin	115	91.3	7.8	0.0	0.9
Levofloxacin	118	88.1	4.2	0.0	7.6
Linezolid	108	88.0	8.3	0.0	3.7
Oxacillin	89	93.3	3.4	2.2	1.1
Penicillin	108	92.6	2.8	3.7	1.0

*Revised to include additional agents.

INTRODUCTION

Automated instrumentation in the clinical microbiology laboratory for the identification and susceptibility testing of bacterial pathogens is in widespread use due to convenience, accuracy and rapidity over that of traditional methods. Databases, database management and use of expert systems are common to most all systems and are periodically updated to expand identification abilities and for detection of newly emerging resistance profiles and mechanisms. Although no system is infallible, all should be reliable, especially with traditional pathogens, and minimize the number of repeat and off-line tests which may greatly lengthen time to reporting of results.

Clinical laboratories have the expectation that automated systems should be able to readily differentiate the most commonly occurring Gram-positive and -negative bacteria and perform reliable susceptibility testing. The over-arching goals of automation are to reduce turn-around times, increase efficiencies and, hopefully, reduce costs.

In addressing clinical needs, the primary goals of accurate bacterial identification and susceptibility testing are to provide healthcare providers with information that can be used to assist in selection of appropriate empiric therapy, assess prognosis and possible public health significance of the offending pathogen/resistance mechanism. Accurate identifications for the laboratorian allow us to properly interpret subsequent antimicrobial susceptibility testing results based upon consensus breakpoints.

Prior studies have demonstrated individual organism-antimicrobial agent weaknesses in automated systems when performing antimicrobial susceptibility testing. In this study we evaluated the accuracy of the Sensititre ARIS 2X (TREK Diagnostic Systems) and Vitek 2 (bioMérieux) systems compared with existing laboratory methods that included primarily the Phoenix (Bectin-Dickenson), but also disk diffusion and E-test (bioMérieux, Inc.) on a contemporary (2010) challenge collection of Gram-positive pathogens recovered at a centralized laboratory serving a regional healthcare network.

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MATERIALS & METHODS

Bacterial isolates. Clinically significant challenge pathogens (162 total) were collected prospectively from both inpatients and outpatients attending a large USA midwestern regional medical center clinic or hospital in 2010 and included staphylococci (98; 53 *Staphylococcus aureus* and 45 other staphylococcal species), *Enterococcus* spp. (52), and streptococci (12; all Lancefield Group B). Specimen sources included skin and soft tissue (58.0%), urine (30.9%), blood (5.6%), respiratory tract (3.7%) and urogenital (1.9%). Details of numbers of isolates and antimicrobial agents tested during this period are found in Tables 1 and 2.

Identification and susceptibility testing methods. All isolates were tested with existing laboratory biochemical algorithms (the Phoenix system, rapid spot tests, biochemical strip tests, traditional tubed biochemicals) with confirmation, where needed, by alternative methods (Sensititre GPID panels incubated and read by the ARIS 2X System; and/or Vitek 2 GP identification panels incubated and read by the Vitek 2 instrument according to the manufacturers' recommendations).

Susceptibility testing was performed using Sensititre susceptibility plates (Part No. GPALL1F) and Vitek 2 susceptibility panels (GP67) and were compared with the existing algorithm ('standard method') consisting of the Phoenix PMIC-106 panel (88.1% of results), disk diffusion using CLSI M100-S20 breakpoints (11.0%) and Etest (for Group B streptococcus only; 0.8%). Consensus was defined as ≥2 of the three test systems with matching categorical results. Acceptability of errors was assessed as described previously (Jorgensen, 1993; CLSI, 2008); generally, total error should be kept to <10%, very major error to <1.5% and major error to <3%.

Table 1. Summary of susceptibility testing agreement and errors by antimicrobial agent when testing Gram-positive isolates (staphylococci, enterococci and streptococci) comparing data generated by the three automated systems only.

			% Error Rate			
Antimicrobial Agent	**n**	**% Agreement All Systems**	**Phoenix**	**Sensititre ARIS 2X**	**Vitek 2**	**No Agreement Between Methods**
Ampicillin	52	94.2	1.9	0	3.8	0
Cefoxitin Screen	90	77.8	7.8	13.3	1.1	0
Clindamycin	101	91.1	1.0	3.0	2.0	1
Erythromycin	115	91.3	7.8	0	0.9	0
Gentamicin	82	100	0	0	0	0
Levofloxacin	118	88.1	4.2	0	7.6	0
Linezolid	108	88.0	8.3	0	3.7	0
Nitrofurantoin	126	96.0	0	0.8	2.4	0
Oxacillin	89	93.3	3.4	2.2	1.1	0
Penicillin	108	92.6	2.8	3.7	1.0	0
Tetracycline	109	96.3	0.9	0.9	1.8	0
Trimethoprim/Sulfamethoxazole	55	100	0	0	0	0
Vancomycin	146	99.3	0	0	0	1
Total	**1299**	**92.8**	**3.0**	**1.8**	**2.0**	**2 (0.2%)**

RESULTS

- A total of 3,897 categorical interpretations were generated for up to 13 antimicrobial agents when testing 162 contemporary Gram-positive pathogens by three susceptibility test systems.
- Overall agreement between the 3 systems was 92.8%; only two organism-antimicrobial agent combinations (0.2%) failed consensus (categorical agreement by ≥2 of the three test systems; Table 1).
- Total error rates were 3.2%, 1.8% and 2.0% for the standard method (3.0% for Phoenix), Sensititre ARIS 2X and Vitek 2 automated systems, respectively, well below recognized thresholds.
- Rates for very major (VME), major (ME) and minor (MiE) errors for Sensititre were, respectively, 1.5%, 0.1% and 0.2%; for Vitek 2, 0.3%, 0.4% and 1.3%; and for the standard method, 0.2%, 1.5% and 1.5%, all very acceptable.
- VME were detected with the standard method when testing the combinations of staphylococcus/penicillin (2.3%) and Group B streptococcus/clindamycin (9.1%); ME with staphylococcus/linezolid (13.5%) and Group B streptococcus/clindamycin (9.1%); and MiE with staphylococcus/erythromycin (7.5-12.2%), enterococcus/linezolid (14.3%)/levofloxacin (9.5%).
- Sensititre VME occurred with combinations of staphylococcus/penicillin (9.3%)/clindamycin (8.1%); and MiE with enterococcus/nitrofurantoin (12.7%).
- Vitek 2 VME occurred when testing streptococcus /clindamycin (18.2%); and MiE with staphylococcus/levofloxacin (4.5-13.2%) and enterococcus/linezolid (19.0%).
- Those agents displaying ≥95% agreement among all isolates included gentamicin, nitrofurantoin, tetracycline, trimethoprim/sulfamethoxazole and vancomycin.
- Error rates for the cefoxitin screen when testing coagulase negative staphylococci should be discounted as this test does not currently apply to CNS.

Table 2. Susceptibility testing summary for 162 Gram-positive bacterial pathogens (all year 2010) when tested by two automated systems and the standard laboratory method (includes results from the Phoenix [88.1%], but also disk diffusion [11.1%] and Etest [Group B streptococci, 0.8%]).

			% Error Rate											
			Very Major Error			Major Error			Minor Error			Combined Error		
Isolate Species/Group	**Antimicrobial Agent**	**n**	**Std. Method**	**Sensititre ARIS 2X**	**Vitek 2**	**Std. Method**	**Sensititre ARIS 2X**	**Vitek 2**	**Std. Method**	**Sensititre ARIS 2X**	**Vitek 2**	**Std. Method**	**Sensititre ARIS 2X**	**Vitek 2**
Staphylococcus aureus (n=53)	Cefoxitin Screen	52	0	0	0	1.9	0	1.9	0	0	0	1.9	0	1.9
	Clindamycin	53	0	0	0	0	0	0	0	0	0	0	0	0
	Erythromycin	53	0	0	0	0	0	0	7.5	0	1.9	7.5	0	1.9
	Gentamicin	51	0	0	0	0	0	0	0	0	0	0	0	0
	Levofloxacin	53	0	0	0	0	0	0	0	0	13.2	0	0	13.2
	Linezolid	50	0	0	0	0	0	0	0	0	0	0	0	0
	Nitrofurantoin	51	0	0	0	0	0	0	0	0	0	0	0	0
	Oxacillin	53	0	0	0	1.9	1.9	1.9	0	0	0	1.9	1.9	1.9
	Penicillin	53	1.9	0	0	0	0	0	0	0	0	1.9	0	0
	Tetracycline	52	0	0	0	0	0	0	0	0	0	0	0	0
	Trimethoprim/Sulfamethoxazole	52	0	0	0	0	0	0	0	0	0	0	0	0
	Vancomycin	52	0	0	0	0	0	0	0	0	0	0	0	0
	Total	**625**	**0.2**	**0**	**0**	**0.3**	**0.2**	**0.3**	**0.6**	**0**	**1.3**	**1.1**	**0.2**	**1.6**
Other *Staphylococcus* spp. (n=45)	Cefoxitin Screen	38	0	0	0	15.8	0	0	0	0	0	15.8	31.6	0
	Clindamycin	37	0	8.1	0	0	0	0	2.7	0	0	2.7	8.1	0
	Erythromycin	41	0	0	0	0	0	0	12.2	0	0	12.2	0	0
	Gentamicin	31	0	0	0	0	0	0	0	0	0	0	0	0
	Levofloxacin	44	0	0	0	0	0	0	4.5	0	4.5	4.5	0	4.5
	Linezolid	37	0	0	0	13.5	0	0	0	0	0	13.5	0	0
	Nitrofurantoin	38	0	0	0	0	0	0	0	0	2.6	0	0	2.6
	Oxacillin	36	0	2.8	0	5.6	0	0	0	0	0	5.6	2.8	0
	Penicillin	43	2.3	9.3	2.3	2.3	0	0	0	0	0	4.7	9.3	2.3
	Tetracycline	38	0	0	2.6	0	0	0	0	2.6	2.6	0	2.6	5.3
	Trimethoprim/Sulfamethoxazole	3	0	0	0	0	0	0	0	0	0	0	0	0
	Vancomycin	42	0	0	0	0	0	0	0	0	0	0	0	0
	Total	**428**	**0.2**	**4.7**	**0.5**	**3.3**	**0**	**0**	**1.9**	**0.2**	**0.9**	**5.4**	**4.9**	**1.4**
Streptococcus, Group B (n=12)	Clindamycin	11	9.1	0	18.2	9.1	0	0	0	0	0	18.2	0	18.2
	Penicillin	12	0	0	0	0	0	0	0	0	0	0	0	0
	Total	**23**	**4.3**	**0**	**8.7**	**4.3**	**0**	**0**	**0**	**0**	**0**	**8.7**	**0**	**8.7**
Enterococcus species (n=52)	Ampicillin	52	0	0	0	1.9	0	3.8	0	0	0	1.9	0	3.8
	Erythromycin	21	0	0	0	0	0	0	0	0	0	0	0	0
	Levofloxacin	21	0	0	0	4.8	0	0	9.5	0	0	14.3	0	0
	Linezolid	21	0	0	0	4.8	0	0	14.3	0	19.0	19.0	0	19.0
	Nitrofurantoin	37	0	0	0	0	0	2.7	2.7	12.7	2.7	2.7	2.7	5.4
	Tetracycline	19	0	0	0	0	0	0	5.3	0	0	5.3	0	0
	Vancomycin	52	0	0	0	0	0	0	0	0	0	0	0	0
	Total	**223**	**0**	**0**	**0**	**1.3**	**0**	**1.3**	**3.1**	**0.4**	**2.2**	**4.5**	**0.4**	**3.6**
	Total Error	**1299**	**0.2**	**1.5**	**0.3**	**1.5**	**0.1**	**0.4**	**1.5**	**0.2**	**1.3**	**2.8**	**0.9**	**2.1**

CONCLUSIONS

- A total of 3,897 categorical interpretations were generated for 13 antimicrobial agents when testing 162 contemporary Gram-positive pathogens; only two organism-antimicrobial agent combinations (0.2%) failed consensus (categorical agreement by ≥2 of the three test systems).
- Overall, acceptable categorical agreement was achieved between the three systems (92.8%) with only slight total error differences detected (standard method 3.2% [Phoenix, 3.0%] > Vitek 2, 2.0% > Sensititre ARIS 2X, 1.8%).
- VME, ME and MiE errors for Sensititre ARIS 2X were, respectively, 1.5%, 0.1% and 0.2%; for Vitek 2, 0.3%, 0.4% and 1.3%; and for the standard method, 0.2%, 1.5% and 1.5%, all within acceptable limits.
- Most errors occurred when testing the combinations of staphylococci with penicillin and clindamycin; Group B streptococci with clindamycin; and enterococci with linezolid.
- Comparative data using frozen-form microbroth dilution panels remains to be performed to adjust final error rate analyses.
- While limitations with each system were apparent and require consideration with implementation depending upon the patient population being served and prevalence of commonly occurring pathogens, the automated systems provided a high level of concordance and very low total error rates.

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