



## Bacteriology

# Comparison of Sensititre microdilution method to other standard methods for susceptibility testing of coagulase-negative staphylococci from paediatric blood cultures☆☆☆

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## ABSTRACT

Susceptibility testing on 60 isolates of *Staphylococcus epidermidis* and 20 isolates of *Staphylococcus hominis* was performed using the Sensititre (Trek Diagnostics, Cleveland, OH, USA) method and compared with Vitek-2 automated susceptibility testing (bioMérieux, Marcy l'Etoile, France), MICE strips for minimum inhibitory concentration determination (Oxoid, Basingstoke, UK), and disc diffusion (MAST, Bootle, UK). The categorical agreement between Vitek and Sensititre was greater than 90% for agents tested against *S. epidermidis* except for tetracycline (52%) and oxacillin (67%), and for *S. hominis*, the agreement was greater than 90% except for tetracycline (80%), gentamicin (65%), and oxacillin (75%). The categorical agreement between disc diffusion and Sensititre was greater than 90% for antimicrobials tested against *S. epidermidis* except for tetracycline (45%), moxifloxacin (70%), clindamycin (88%), and cefoxitin (80%), while against *S. hominis*, the categorical agreement was greater than 90% for antimicrobials tested except for gentamicin (65%), tetracycline (75%), moxifloxacin (80%), and ciprofloxacin (75%). Categorical agreement between MICE strips and Sensititre for *S. epidermidis* and *S. hominis* was greater than 95% for vancomycin and daptomycin. Sensititre method shows good categorical agreement with other standard methods for susceptibility testing of *S. hominis* and *S. epidermidis*.

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## 1. Introduction

The interpretation of susceptibility results is critical as they are used by clinicians to guide antimicrobial therapy. Minor differences, particularly for antimicrobial agents that may be reported as intermediate susceptibility, can cause a significant impact for patient care as it can lead to a change in antimicrobial use (Hombach et al., 2013). Sensititre susceptibility testing method is based on micro-wells, which have 2-fold serial dilutions of antimicrobial in each well, which can be used for testing for a range of bacteria and fungi (Pfäller et al., 2012; (Sapino et al., 2012). The benefit of these systems is that they allow for automation of plate reading and, thus, the ability to increase the efficiency of a laboratory, thus leading to cost-savings and facilitating the introduction of automation (Staneck et al., 1988). Recent studies have shown that Sensititre method performs well when compared to other standard methods for the susceptibility

testing of *Candida* spp. and also *Mycobacterium tuberculosis* (Farina et al., 2011; (Hall et al., 2012; (Pfäller et al., 2012). Sensititre plates have also been shown to correctly identify 68.9% of coagulase-negative staphylococci (CoNS), including correct identification of *Staphylococcus epidermidis* in 73% of cases (Garza-Gonzalez et al., 2010).

CoNS causes important morbidity in hospitalised paediatric patients as it is responsible for the majority of central line-associated bloodstream infection (Dimitriou et al., 2011). It is therefore important to rely on prompt and accurate sensitivity results to guide antimicrobial therapy. The purpose of this study was to examine the categorical agreement that existed between Sensititre and 3 other established antimicrobial susceptibility methods when using British Society of Antimicrobial Chemotherapy (BSAC) guidelines for CoNS recovered from positive paediatric blood cultures. The 3 other methods examined were disc diffusion (MAST Diagnostics, UK), MICE strips for determination of MIC (Oxoid, UK), and Vitek-2 (bioMérieux, France) automated susceptibility testing.

## 2. Materials and methods

The study was performed in the Microbiology Laboratory of Alder Hey Children's NHS Foundation Trust. Frozen (−80 °C) isolates of CoNS grown from positive blood cultures samples between January 2011 and February 2012 were recovered from storage beads.

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**Table 1**Comparison of susceptibility testing performed on the 60 isolates of *S. epidermidis* by disc diffusion, Vitek, and Sensititre method.

<i>S. epidermidis</i>	Sensititre					Disc diffusion				
	CA	% CA	MajD	VMajD	Minor	CA	%CA	MajD	VMajD	Minor
Vancomycin	59	98%		1						
Gentamicin	58	97%	2			58	97%		2	
Linezolid	60	100%				58	97%	2		
Rifampicin	59	98%	1			56	93%			4
Synercid	60	100%				59	98%	1		
Tetracycline	31	52%	1	1	27	20	33%		2	38
Erythromycin <sup>a</sup>	53	90%	2		4	58	97%	2		
Penicillin	57	95%	1	2		59	98%		1	
Clindamycin	56	93%	1		3	57	95%	3		
Ciprofloxacin <sup>a</sup>	57	97%	1	1		59	98%			
Oxacillin	40	67%	1	19						
Disc Diffusion										
Gentamicin	56	93%	4							
Linezolid	58	97%		2						
Rifampicin	55	92%	2	3						
Synercid	59	98%		1						
Tetracycline	27	45%	4	4	25					
Erythromycin <sup>a</sup>	55	93%	1		4					
Penicillin	56	93%	2	2						
Moxifloxacin	42	70%	2	1	15					
Clindamycin	53	88%	1	3	3					
Ciprofloxacin <sup>a</sup>	57	97%	1	1						
Cefoxitin	48	80%	2	8	2					

<sup>a</sup> Fifty-nine isolates only.

Identification of the CoNS species was performed using the Vitek-2 Gram-positive card (bioMérieux, France). Disc diffusion was performed in accordance with the most recent BSAC guidelines (Andrews and Howe, 2011). The antimicrobials tested by disc diffusion were gentamicin, linezolid, rifampicin, quinupristin-dalfopristin, tetracycline, erythromycin, penicillin, clindamycin, and ciprofloxacin. Plates were incubated at 35–37 °C for 20–24 hours. For vancomycin and daptomycin, MIC strips (Oxoid, UK) were used to determine the MIC as disc diffusion was not possible in accordance with BSAC guidance.

The Sensititre method was performed in line with the manufacturer's instructions. In brief, a pure culture of the CoNS isolate, which had been grown overnight on blood agar, was used with sterile water

to make a 0.5 McFarland turbidity suspension. The Sensititre plate (Code GPALL1F; Trek Diagnostics, USA) consisted of serial 2-fold dilutions of the following antimicrobials, vancomycin (0.25–32 mg/L), gentamicin (2–16 mg/L), linezolid (1–8 mg/L), rifampicin (0.5–4 mg/L), quinupristin-dalfopristin (0.5–4 mg/L), tetracycline (2–16 mg/L), erythromycin (0.25–4 mg/L), penicillin (0.06–8 mg/L), clindamycin (0.5–2 mg/L), ciprofloxacin (1–2 mg/L), daptomycin (0.5–4 mg/L), and oxacillin with 2% sodium chloride (0.25–4 mg/L). From this suspension, 10 µL was transferred using a calibrated loop into the Sensititre Muller Hinton broth tube, and then 50 µL was transferred into each well using an 8-channel pipette. The plate was then covered with the adhesive seal that was supplied with the kit and incubated

**Table 2**Comparison of susceptibility testing performed on the 20 isolates of *S. hominis* by disc diffusion, Vitek, and Sensititre method.

<i>S. hominis</i>	Sensititre					Disc diffusion				
	CA	% CA	MajD	VMajD	Minor	CA	%CA	MajD	VMajD	Minor
Vancomycin	19	95%	1							
Gentamicin	13	65%	6	1		20	100%			
Linezolid	20	100%				20	100%			
Rifampicin	19	95%	1			20	100%			
Synercid	20	100%				19	95%	1		
Tetracycline	16	80%			4	15	75%			5
Erythromycin	19	95%			1	20	100%			
Penicillin	19	95%		1		20	100%			
Clindamycin	18	90%		1	1	19	95%		1	
Ciprofloxacin	19	95%	1			16	80%	4		
Oxacillin	15	75%	4	1						
Disc diffusion										
Gentamicin	13	65%	6	1						
Linezolid	20	100%								
Rifampicin	19	95%	1							
Synercid	19	95%		1						
Tetracycline	15	75%			5					
Erythromycin	19	95%			1					
Penicillin	19	95%		1						
Moxifloxacin	16	80%			4					
Clindamycin	19	95%			1					
Ciprofloxacin	15	75%	1	4						
Cefoxitin	19	95%		1						

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