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Use of WalkAway MicroScan system colistin well when determining the susceptibility of *Pseudomonas aeruginosa* and *Acinetobacter baumannii* recent clinical isolates

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Sir,

The increasing number of multiresistant non-fermenter Gram-negative bacilli isolates has boosted the use of antibiotics, like colistin, for the treatment of the infections caused by these microorganisms [1]. Unfortunately, the assessment of the colistin susceptibility is not an easy task in a microbiology laboratory, thus finding a solution for this situation has become essential. It has been described that disc-diffusion and E-test methods are not reliable tools, since a high percentage of false resistance has been detected as their result [2]. Based on the EUCAST guidelines [3], the gold standard for the colistin sensitivity determination would be the broth microdilution method, even though this implies the added difficulty of the manual processing. A few alternatives can be considered, like the Sensititre (ThermoFisher Diagnostics, U.S.) and the MicroScan (Beckman Coulter, U.S.) panels but, even in these, the several performed studies have found that false resistances can appear in up to 65% of the cases [4]. Another useful method, considering the EUCAST's proposal, is the manual microdilution used in UMic test (BioCentric, France), which we are currently using in our laboratory in order to confirm the resistances that the MicroScan system has previously found. The aim of our study was to compare the automatized MicroScan and E-test (gradient diffusion) methods to the UMic manual microdilution.

A total of 23 isolates (17 *Pseudomonas aeruginosa* and 6 *Acinetobacter baumannii*), all proceeding from urine cultures of different clinical episodes, have been studied between April 2018 and April 2019, being all interpreted as colistin-resistant by the MicroScan system (table 1). Focusing our attention on the study of false positives (*major errors*), colistin suscep-

tibility has been evaluated using the gradient-diffusion E-test (MIC Strip, Liofilchem, Italy) method in Müller-Hinton agar (BD, Spain) and through microdilution (UMic), which was used as the reference method. The colistin susceptibility study was carried out according to the manufacturers' instructions. The ATCC 27853 (American Type Culture Collection) strain of *Pseudomonas aeruginosa* was used as control.

The possible MIC ranges were ≤ 0.06 to > 64 mg/L for the manual microdilution, ≤ 2 to > 4 mg/L for the MicroScan system and ≤ 0.016 to > 256 mg/L corresponding to the E-test method. The obtained results are presented in the table 1. A MIC > 2 mg/L was obtained in 7 isolates (30.4% of the samples) through E-test method, in 4 isolates (17.4%) through manual microdilution and in 2 isolates (8.7%) using both techniques. A total of 83% of the isolates that MicroScan system identified as having a MIC ≥ 4 mg/L resulted to be susceptible (MIC ≤ 2 mg/L) after using the reference method of our laboratory, therefore meaning that the MicroScan system possesses a very low specificity. Concomitantly, the E-test method resulted not to be efficient at determining this possible resistance since its outcomes were MIC > 2 mg/L in isolates that were susceptible performing microdilution and, at the same time, it showed sensitivity in resistant isolates (false negative or *very major errors*).

The gathered data brings into question the utility of the colistin wells in these panels from MicroScan system, although maybe their negative predictive value justifies their presence. Therefore, we think that the colistin well should be reviewed in order to determine the possibility of being improved, removed or reinterpreted in the manufacturer's instructions.

For the moment, our recommendation when colistin resistance appears in non-fermenter Gram-negative bacilli, is to always perform a second interpretation, using UMic or other method, following the EUCAST's suggestions. Larger studies involving a higher number of isolates are necessary in order to reach global reliable conclusions.

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Table 1 Susceptibility to colistin in *Pseudomonas aeruginosa* and *Acinetobacter baumannii* clinical isolates.

Isolates	Microorganisms	MicroScan System		E-Test System		UMic System	
		MIC (mg/L)	S/R	MIC (mg/L)	S/R	MIC (mg/L)	S/R
1	<i>A. baumannii</i>	> 4	R	2	S	0.06	S
2	<i>A. baumannii</i>	> 4	R	2	S	0.125	S
3	<i>A. baumannii</i>	4	R	2	S	0.5	S
4	<i>A. baumannii</i>	4	R	2	S	0.5	S
5	<i>P. aeruginosa</i>	> 4	R	1.5	S	0.5	S
6	<i>P. aeruginosa</i>	4	R	1	S	1	S
7	<i>P. aeruginosa</i>	4	R	1.5	S	1	S
8	<i>P. aeruginosa</i>	4	R	1	S	1	S
9	<i>A. baumannii</i>	4	R	0.75	S	1	S
10	<i>P. aeruginosa</i>	4	R	1.5	S	1	S
11	<i>P. aeruginosa</i>	> 4	R	1	S	1	S
12	<i>P. aeruginosa</i>	4	R	3	R	1	S
13	<i>P. aeruginosa</i>	> 4	R	1	S	1	S
14	<i>P. aeruginosa</i>	4	R	2	S	1	S
15	<i>P. aeruginosa</i>	> 4	R	6	R	1	S
16	<i>P. aeruginosa</i>	4	R	6	R	1	S
17	<i>P. aeruginosa</i>	> 4	R	0.75	S	2	S
18	<i>P. aeruginosa</i>	> 4	R	3	R	2	S
19	<i>P. aeruginosa</i>	4	R	3	R	2	S
20	<i>P. aeruginosa</i>	4	R	1.5	S	4	R
21	<i>P. aeruginosa</i>	4	R	4	R	4	R
22	<i>A. baumannii</i>	> 4	R	24	R	32	R
23	<i>P. aeruginosa</i>	> 4	R	2	S	32	R

MIC= minimum inhibitory concentration.

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None to declare.

CONFLICTS OF INTEREST

The authors declare that they have no conflicts of interest

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