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O0747 Multi-laboratory validation of the rapid method for antimicrobial susceptibility testing directly from positive blood cultures proposed by EUCAST

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Background: Rapid antimicrobial susceptibility testing (RAST) directly from positive blood cultures (BC) can shorten the time to antibiotic susceptibility reports. EUCAST is currently developing a standardized RAST method (Poster 165, ECCMID 2017). Here the proposed method was evaluated in 36 laboratories under routine conditions.

Materials/methods: Each laboratory was asked to perform the RAST method on 40 consecutive positive BC, 20 with Gram-negative and 20 with Gram-positive bacteria. Inhibition zones for antibiotics commonly used in the treatment of blood stream infections were measured after 4, 6 and 8h incubation and interpreted according to breakpoints developed to match the 4, 6 and 8h readings. Two BC systems (BD and bioMérieux) and media/disks from 4 manufacturers were used. All isolates were sent to the EUCAST development laboratory for disk diffusion according to EUCAST standard methodology. In this evaluation, the number of inhibition zones possible to read and the categorical agreement vs. standard disk diffusion using the tentative breakpoints for early reading were evaluated for *Staphylococcus aureus* (n=242) and *Escherichia coli* (n=386).

Results: After 4, 6 and 8h incubation, zone diameters for *S.aureus/E.coli* could be read for 65%/91%, 92%/99% and 95%/99% of the isolates. The number of categorical errors was low (Table 1) and there was no systematic difference between the BC systems. There were few (≤0.5%) false susceptible results (VME), but the number of resistant isolates in the collection was low. False resistance (ME) was most prominent after 4h incubation, especially for *S.aureus*, but the number decreased as incubation continued. This was also true for the number of results categorized as ATU (Area of Technical Uncertainty). These were mainly related to piperacillin-tazobactam in *E.coli*. When the 4h *S.aureus* reads were excluded, 86% and 97% of the laboratories, respectively, reported ≥97% and ≥95% of all readings correctly.

Conclusions: The proposed EUCAST rapid AST methodology and breakpoints for *S.aureus* and *E.coli* could be implemented in a large number of laboratories, independent of the BC system used. The high proportion of isolates ending up in ATU for piperacillin-tazobactam and the false resistance observed in *S.aureus* after 4 h incubation will be looked into further.

Table 1

Number of readings with EUCAST RAST methodology and categorical agreement with standard AST

	***S. aureus* (n=242)** Cefoxitin, norfloxacin, erythromycin, gentamicin			***E. coli* (n=386)** Cefotaxime, ceftazidime, piperacillin-tazobactam, meropenem, ciprofloxacin, amikacin, tobramycin, gentamicin			***E. coli* (n=386)** Piperacillin-tazobactam excluded		
Incubation time	**4h**	**6h**	**8h**	**4h**	**6h**	**8h**	**4h**	**6h**	**8h**
Number of possible tests[a]	968	968	968	3 088	3 088	3 088	2 702	2 702	2 702
Number of performed tests[b]	952	956	892	3 034	3 027	2 768	2 651	2 645	2 419
Number of zones registered[c]	623	880	844	2 756	2 993	2 752	2 415	2 613	2 404
	Categorical agreement (%)			**Categorical agreement (%)**					
Correct	66	92	95	77	81	84	88	93	95
mE	0.0	0.0	0.0	0.3	0.1	0.1	0.2	0.2	0.1
ME	8.5	0.3	0.4	1.6	0.4	0.2	1.8	0.5	0.3
VME	0.2	0.3	0.5	0.1	0.1	0.1	0.1	0.1	0.1
ATU	25	7.2	4.0	20	18	16	10	6.2	4.0

[a]Number of possible tests = Total number of possible isolate-agent combinations

[b]Number of performed tests = Number of possible tests after excluding missing data (e.g. disk forgotten or laboratory opening hours too short)

[c]Number of zones registered = Number of performed tests with readable inhibition zones

mE (minor Error) = Categorised as susceptible (S) or resistant (R) with RAST when intermediate (I) with standard method.

ME (Major Error) = False resistant

VME (Very Major Error) = False susceptible

ATU (Area of technical uncertainty) = RAST results close to the breakpoint, judged not reliable to report. These should be read after further incubation or retested with standard method