31st ECCMID Online 9 – 12 July 2021

ESCMID EUROPEAN SOCIETY OF CLINICAL MICROBIOLOGY AND INFECTIOUS DISEASES

A unique pre analytical workflow to achieve identification and AST from positive blood culture with Sepsityper® kit on VITEK MS and VITEK2 instruments

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INTRODUCTION

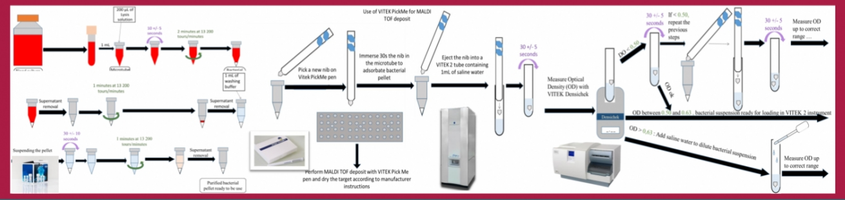
Diagnostic of sepsis remains based on blood culture. However, traditional workflow of positive blood culture includes subculture of colony to perform identification (ID) and antibiotic susceptibility tests (AST) and need in the most of the case 2-3 days for final results. During this time, antimicrobial therapies are only "empiric" guideline based and could be inappropriate due to increased resistant microorganisms. Thus, the analytical blood culture workflow need to be accelerated to obtain ID and AST the same day of the blood culture bottle (BCB) positivity.

AIM

Design and evaluation of an analytical workflow from positive blood culture bottle to perform ID, AST and detection of some resistance mechanisms impacting the empirical treatment of sepsis.

METHOD

- Prospective single center study on 100 positives BCB (BACT/ALERT®) including
  - 43 enterobacterales;
  - 7 non fermenting bacteria with 4 *Pseudomonas aeruginosa*;
  - 2 Gram positive rod with 1 *Corynebacterium afermentans* and 1 *Cutibacterium acnes*;
  - 4 *Staphylococcus aureus*
  - 25 *S. non aureus*; 7 *Streptococcus sp.*; 11 *Enterococcus sp.* and 2 *Candida* yeast.
- The 2 two steps SepsiTyper extraction kit was performed on positive BCB less than 1h after their positivity.
- On the obtained pellet, PickMe® (PM) pen was used to perform slide deposit and analysed by VITEK MS instrument.
- The same PM tip was ejected on saline tube for VITEK 2 (V2) and optical density was measured before loading on the V2 with relevant AST cards according to ID result.
- On the remaining pellet, immune chromatographic tests (ICT) for PBP2a (Clearwiew, Abbott); ESBL (CTX-MM, Eurobio) or carbapenemases (RESIST5, CORIS BioConcept) were also done.
- All data were anonymously compared with standard of care (SOC).



RESULTS

- *Compared to SOC, 85/96 (92%), 3/96 (2%) and 8/96 (8%) BCB identifications were correctly, erroneous and not identified respectively (Table 1).*
- *AST results were done in the Table 2.*
- *Regarding ICT, PBPB2a, CTX-M and carbapenemase tests were performed on 4 S. aureus, 21 E. coli and K. pneumoniae and 6 E. cloacae respectively. Only 1/21 were CTX-M positive whereas all the other ICT were negative in accordance with routine results.*

Table 1 ID results

	ID OK				False ID			No ID	TOTAL
	Test 1	Test 2	Test 3	TOTAL (%)	Test 1	Test 2	TOTAL (%)		
Gram Negative bacilli	43	5	1	49 (100%)	0	0	0	0	49
Gram Positive bacilli	0	0	0	0 (0%)	1	1	2 (100%)	0	2
Staphylococcus	15	3	0	18 (64%)	0	1	1 (4%)	8 (28%)	27
Streptococcus + Enterococcus	13	2	1	16 (100%)	0	0	0	0	16
Yeast	2	0	0	2 (100%)	0	0	0	0	2
TOTAL	73	10	2	85 (89%)	1	2	3 (3%)	8 (8%)	96

Table 2 AST results on V2

	AST errors	Same MIC	+/- 1 MIC	≥ +/- 2 MIC	Same SIR categories	Minor error	Major error	Very major error
Gram negative bacilli - N233+XN12 card	0	91%	5%	4%	98%	1%	0	1%
Non fermenting Gram negative bacilli – N240 card	7%	62%	5%	28%	93%	0	0	0
Staphylococcus sp. – P631 card	3%	68%	7%	22%	83%	5%	4%	5%
Streptococcus sp. / Enterococcus sp. – P606 card	0	54%	0	46%	69%	0	20%	11%

CONCLUSIONS

Using Sepsityper kit for sample preparation, PM pen for deposit and AST inoculum preparation, ICT and VITEKs instruments, this "fast and easy to perform" **consolidated analytical protocol give ID results and main resistance mechanisms in less than 1h followed by AST results in less than 15h without additional or specific instruments**. Additional tests must be perform with more resistant strains to evaluate ICT directly on positive BCB as well as V2 cards. An alternative method could be to replace Vitek AST cards by rapid AST from EUCAST.

ACKNOWLEDGEMENTS

We are grateful to Bruker for the gift of Sepsityper kits. We also thank bioMérieux for the loan of Vitek 2 instrument.

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