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FULL PAPER

Accelerate Pheno™ system in sepsis by Gram-negative pathogens: four months of hospital experience

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SUMMARY

This study reports our experience with the Accelerate Pheno™ system (ACC) to guide management of patients with sepsis by Gram-negative pathogens. A diagnostic workflow, based on pathogen and resistance genes detection or ACC testing, was applied to 33 patients. Clinical and microbiological data were recorded, and analysis of broad-spectrum agents sparing was performed. Antimicrobial susceptibility results by ACC were available for 28 of 33 patients (84.85%). Among 434 microorganism-antimicrobial combinations, categorical agreement was 97.93%, very major errors 0.23%, major errors 1.15%, and minor errors 0.69%. Time to report (mean ± SD) of ACC results was 27.14±6.90 h from sample collection, significantly shorter (p<0.001, Δ = 19.96 h, 95% CI: 24.71-15.22) than that of the standard method (47.10±11.92 h). A switch from empiric to targeted therapy was observed in 14 of 28 patients (50.0%), duration of empiric therapy was 37.73±19.87 h, with a saving of 5.45 piperacillin/tazobactam and 5.28 carbapenems prescribed daily doses. Considering patients in which de-escalation would have been theoretically feasible, 27.69 prescribed daily doses of piperacillin/tazobactam and 19.08 of carbapenems could had been spared, compared to standard methods. In conclusion, ACC could impact positively on the management of septic patients by Gram-negative pathogens.

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INTRODUCTION

The early administration of effective antimicrobials is crucial for septic patient survival (Levy *et al.*, 2018; Kumar *et al*., 2006), but as many as 20-30% of patients receive inadequate antimicrobial treatment (Kumar *et al*., 2009; Ibrahim *et al*., 2000). Moreover, de-escalation of empiric therapy has been shown to be associated with lower mortality (Garnacho-Montero *et al*., 2014).

The challenge of empiric antimicrobial therapy in these patients is the balance between two conflicting objectives: the provision of rapid, effective therapy and the minimization of broad-spectrum antimicrobial use to avoid the emergence of antimicrobial resistance (AMR), which, in turn, hampers the appropriateness of empiric therapy (Pogue *et al*., 2015).

Blood culture (BC) is the reference diagnostic test for pathogen identification (ID) and antimicrobial susceptibility testing (AST) (Levy *et al*., 2018), although with the important limitation of long time-to-result (Opota *et al*., 2015), needing a paradigm shift in diagnostic microbiology for early pathogen ID and AST results (Trotter *et al*., 2019). Different approaches are used to hasten laboratory results in the management of BC. Matrix-assisted laser desorption ionization time-of-flight mass spectrometry (MALDI-TOF MS), directly from positive BC bottles, can provide rapid ID but not AST results; whereas, molecular methods, applied to positive BC, can detect some, but not all, resistance genes. On the other hand, phenotypic AST, which determines minimal inhibitory concentration (MIC) of antimicrobials, is still necessary for tailored, directed treatment (Peker *et al*., 2018). Laboratory automation and 8 h digital reading of plates from positive BC greatly reduces time-to-report (TTR) and shortens the duration of empiric antimicrobial therapy, possibly improving outcome in patients with bloodstream infections (De Socio *et al*., 2018). Recently, the Accelerate Pheno™ system (ACC) has been shown to significantly improve turnaround time in the diagnosis of bloodstream infections (De Angelis *et al*., 2019; Charnot-Katsikas *et al*., 2018)

Few studies have evaluated the clinical impact of these rapid tests. Indeed, the current availability of a large spectrum of rapid diagnostics tests (Trotter *et al*., 2019), makes it mandatory that microbiological diagnosis and treatment rely on specific diagnostic and antimicrobial stewardship programs, based on strict collaboration between clinical and laboratory staff. Diagnostic stewardship is defined as coordinated guidance and interventions to improve appropriate use of microbiological diagnostics to guide therapeutic decisions. It should promote appropriate, timely diagnostic testing and reporting of

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results to guide patient treatment (World Health Organization, 2016).

Italy is characterized by a very high prevalence of multi-drug resistant organisms, a major problem in our hospital as well (De Socio *et al*., 2019), with 32% *Klebsiella pneumoniae* blood isolates producing carbapenemase (Monari *et al*., 2016), 14% *Enterococcus faecium* vancomycin-resistant, and 25% *Staphylococcus aureus* and 90% coagulase-negative staphylococci methicillin-resistant. To address the challenge of timely and appropriate treatment of bloodstream infections, a defined diagnostic workflow to process positive BC, based on local epidemiology and the availability of many rapid tests, was recently designed in our hospital as a part of diagnostic stewardship program and shared with clinicians (*Figure 1*). The ACC test was included in this workflow to provide rapid MIC results, when appropriate, and when other rapid diagnostic tests were not useful.

The aim of this study was to assess how ACC, performed according to predetermined rules, could impact the management of patients with sepsis by Gram-negative pathogens.

MATERIALS AND METHODS

Study design

This prospective observational study was carried out from October 1, 2018 to January 31, 2019 and included patients with sepsis, defined according to the Third International Consensus Definitions for Sepsis and Septic Shock (Singer *et al*., 2016), with positive BC processed according to the workflow described in *Figure 1*.

Blood culture, standard identification and antimicrobial susceptibility testing

Our microbiology laboratory provides diagnostic services to the 800-bed General Hospital of Perugia, Italy, serving a population of around 200,000 people. It operates from 08:00 a.m. to 08:00 p.m., Monday to Friday, and from 08:00 a.m. to 02:00 p.m. on Saturday and Sunday/holidays. Blood cultures were delivered from wards within one hour of collection and incubated immediately 24/7, taking advantage of satellite incubators when the laboratory was closed. Blood cultures were received inoculated into BD BACTEC Plus Aerobic/F and BD BACTEC Lytic/10 Anaerobic/F bottles (Becton Dickinson, Sparks, MD, USA). Positive BC were processed as previously described (De Socio *et al*., 2018). For positive BC processed during laboratory hours 8:00 a.m. - 11:00 a.m., Monday to Friday, ID and AST were set up after 8-h incubation and final results were reported the following day. Outside of these laboratory hours, ID and AST were set up after 18-h incubation, and final results were reported the following day.

Identification of isolates was performed by using the Bruker MALDI Biotyper instrument (Bruker Daltonik GmbH, Bremen, Germany) as described elsewhere (Leli *et al*., 2013). Antimicrobial susceptibility testing was performed with the BD Phoenix (Becton Dickinson) automatic system. For bacterial isolates suspected to be multi-drug resistant, AST was carried out with broth microdilution using lyophilized custom plates (MICRONAUT Merlin, Bornheim-Hesel, Germany, ISO 2776-1 standard inoculum) and interpreted according to current EUCAST clinical breakpoints (European Committee on Antimicrobial

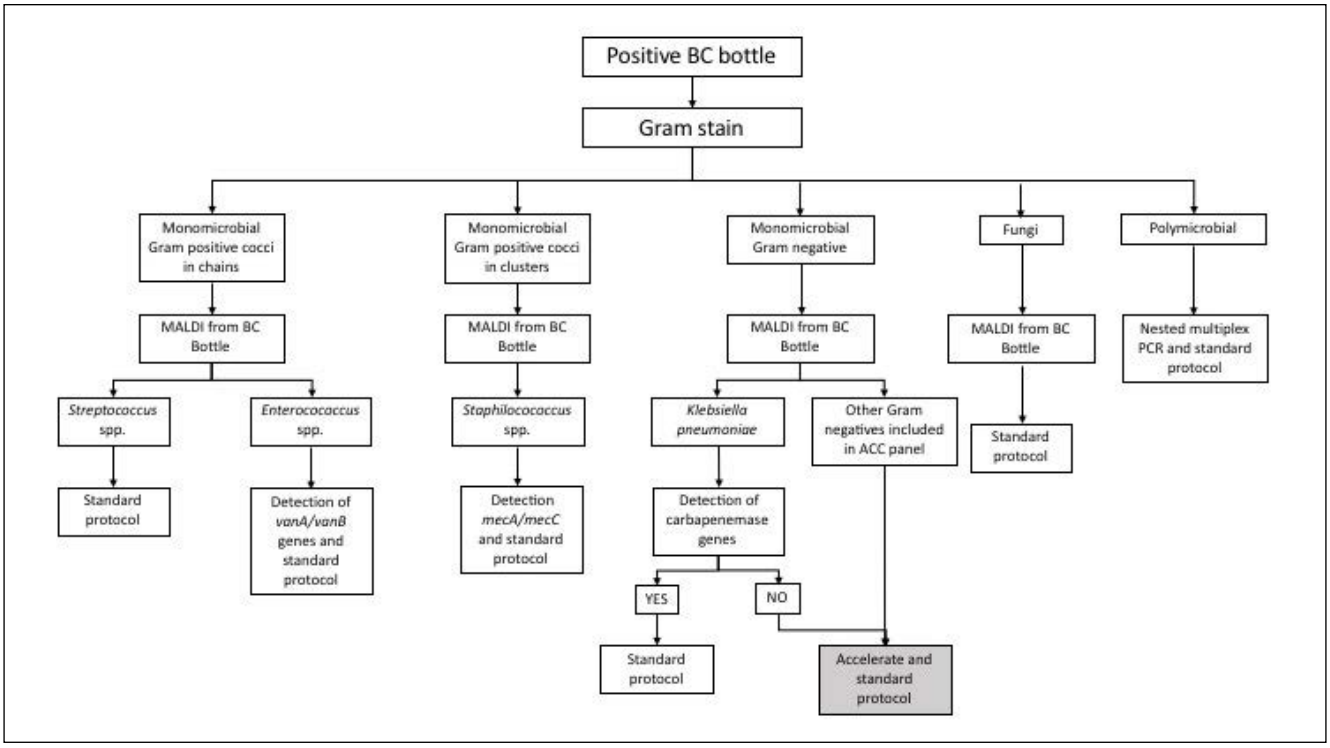

Figure 1 - *Workflow used for ACC and standard method testing. For Gram-positive cocci, both the standard method and rapid molecular tests for resistance genes detection were used. ACC was used for all Gram-negative organisms included in ACC panel, except carbapenemase-producing Klebsiella pneumoniae isolates. Organisms not included in ACC panel were analyzed only by standard protocol. Standard protocol: ID and AST from colonies on agar plates sub-cultured from positive BC.*

Susceptibility Testing, 2018; European Committee on Antimicrobial Susceptibility Testing, 2019).
The Xpert Carba-R assay (Cepheid, Sunnyvale, CA, USA) and/or the immunochromatographic test NG-Test CARBA5 (NG Biotech, Guipry, France) from BC bottles were used for rapid detection of carbapenemase producing *Enterobacteriaceae*. Molecular detection of *mecA/mecC* and *vanA/vanB* genes was done by the GeneXpert system (Cepheid). For polymicrobial BC the BC FilmArray assay (bioMérieux, Marcy l'Etoile, France) was used. All molecular tests were performed according to the manufacturers' instructions.

Accelerate Pheno™ System
BC specimens (1 mL) were tested using ACC within 8 h of BC positivity with the Accelerate PhenoTest™ BC kit (Accelerate Diagnostics, Tucson, AZ, USA), according to the manufacturer's instructions. The analysis software Accelerate Diagnostics host application version 1.3.2 was used. Only one positive BC (aerobic or anaerobic bottle) per patient was included in the study.

Evaluation of accuracy of ID and AST results obtained by Accelerate Pheno™
For the same organism, ID and AST results obtained using ACC were compared to those of standard protocol. Identification results were classified as 'correct' (genus or species level), 'no identification', or 'incorrect'. To compare AST results, MIC values were converted to clinical categories (S/I/R) according to current EUCAST criteria (European Committee on Antimicrobial Susceptibility Testing, 2018; European Committee on Antimicrobial Susceptibility Testing, 2019), and categorical agreement (CA), very major errors (VME, false susceptibility), major errors (ME, false resistance), and minor errors (mE, susceptible/resistance versus intermediate susceptibility) were assessed. The microdilution method was used to resolve discrepancies.

Data collection
For each patient included in the study, demographic, clinical, and laboratory data were recorded on a Microsoft Excel spreadsheet. Data on antimicrobial therapy were also collected, with specific attention to type and time of empiric therapy, and time to switch to targeted therapy.
Times were recorded using the Epicenter microbiology software package (Becton Dickinson) and the TD-Synergy Laboratory Information System (LIS, Siemens, Italy). Time-to-report was defined as the period of time (h) between BC collection and the availability of AST results, using either the standard or ACC method.

Real and hypothetical impact on therapeutic decisions of Accelerate Pheno™ AST results
The impact on therapeutic decisions using ACC results was evaluated by comparing therapy before (empiric therapy) and after (targeted therapy) the reporting of ACC AST results. The following definitions were used:
1) maintenance of empiric therapy was defined as no change in antimicrobial therapy (one or more antimicrobial drugs), irrespective of its appropriateness based on microbiological results;
2) switch to targeted therapy was defined as de-escalation (discontinuation of one or more broad-spectrum antimicrobials and/or replacement by a narrower-spectrum one) or escalation (addition of narrower-spectrum antimicrobial) therapy, in accordance with AST of the isolated pathogen.

Broad-spectrum antimicrobials sparing was defined as the number of spared prescribed daily doses (PDD) of broad-spectrum antibiotics when comparing results from ACC to standard methods. The hypothetical impact was calculated by assuming that the switch from empiric to targeted antimicrobial therapy was made at the time the ACC AST results were reported. Every effort was made to carefully identify cases in which it was possible to switch from broad-spectrum to narrow-spectrum antibiotic therapy: for each patient, microbiological data (i.e., bacterial isolates from all clinical specimens collected), laboratory parameters (e.g., procalcitonin, blood cell counts, C-reactive protein, renal and liver function, etc.), and the ongoing antimicrobial therapy were reviewed in conference by the investigators (infectious disease specialists, pharmacologists, and clinical microbiologists), and the theoretical feasibility of de-escalation therapy was evaluated.

Statistical analysis
Standard descriptive statistics were used to summarize data, such as mean, standard deviation (SD), median, inter-quartile range (IQR), and percentage. Student's *t*-test was used to assess differences in continuous variables. The time was analyzed and reported as hours in decimal format. A p-value <0.05 was considered statistically significant. MedCalc statistical package, version 18.2.1 (MedCalc software, Ostend, Belgium) was used for all statistical analyses.

Ethics statement
Not required. Samples were collected and results were delivered to wards as part of standard care. Data included in the database were de-identified before access. No personal information was stored in the study database.

Table 1 - *Gram-negative pathogens isolated from monomicrobial positive blood cultures (BC) during the study period.*

Bacterial species	*BC*	*BC suitable for ACC testing*	*BC tested by ACC*
Escherichia coli	24	24	18
Klebsiella pneumoniae	12	6	3
Pseudomonas aeruginosa	4	4	4
Enterobacter spp	4	4	2
Serratia marcescens	4	4	3
Proteus spp	2	2	1
Klebsiella oxytoca	2	2	0
Citrobacter spp	2	2	2
Others not included in ACC panel	13	0	0
Total	**67**	**48**	**33**

RESULTS

Patient population and bacterial isolates

During the study period, 4,998 BC sets were collected from 1,952 patients. Among these, 529 (10.58%) BC sets from 155 patients were positive for one pathogen. Samples containing 79 Gram-positive bacteria and 9 yeasts were not tested by ACC according to the workflow process described in Figure 1. Sixty-seven BC were positive for Gram-negative pathogens, 48 of which were suitable for ACC testing. Among these, 15 could not be tested due to laboratory operating hours (interval time between positivity detection and initiation of ACC testing >8 h), and 33 were ultimately tested (*Table 1*).

The main characteristics of the study population are summarized in *Table 2*.

Table 2 - *Characteristics of the study population.*

Variable	*Value*
Patients	33
Years, median (IQR)	73 (69-80)
Men	15 (45.45)
Hospital ward	
Medicine	14 (42.42)
Hematology/Bone Marrow Transplantation	6 (18.18)
Surgical	4 (12.12)
Intensive Care	4 (12.12)
Emergency Department	5 (15.15)
Laboratory parameters	
Leucocytes, cells x 10^3/mL, Median (IQR)	12.15 (3.14-16.57)
Leukocytosis, >12 x10^3/mL	20 (60,6)
Leucopenia, <4 x 10^3/mL	10 (30.3)
Neutrophils percentage, Median (IQR)	87.8 (79.22-92.28)
CRP, mg/dL, Median (IQR)	8.35 (5.0-17.2)
PCT, ng/mL, Median (IQR)	27.02 (3.25-47.35)
Lactate, mM/L, Median (IQR)	2,65 (1.1 – 4.5)
Clinical data	
Body Temperature, °C, Median (IQR)	38.25 (37.3-38.6)
Temperature >38°C	17 (51.52)
Temperature <36°C	6 (18.18)
Heart Rate, beats/min, Mean ± SD	96.5 ±19.14
Mean Arterial Pressure, mmHg, Mean ± SD	75.17 ±18.62
Septic Shock	9 (27.27)
Sequential Organ Failure Assessment Score, Median (IQR)	7.00 (5.00-10.00)
30-Days Mortality	10 (30.30)
Concomitant diseases	
Hypertension	18 (54.55)
History of cardiovascular disease	13 (39.39)
Chronic renal failure	12 (36.36)
Malignancy	12 (36.36)
Diabetes	9 (27.27)
Chronic lung disease	8 (24.24)
Dyslipidemia	5 (15.15)
Chronic liver disease	3 (9.09)
Dementia	2 (6.06)

Data were obtained at the time of blood culture draw, with the exception of mortality.
Values are number (%), unless otherwise specified.
CRP, C-reactive protein.
PCT, procalcitonin.

Evaluation of accuracy of ID and AST results obtained by Accelerate Pheno™

Samples were analyzed to compare the ID and AST results obtained from the same positive BC after standard method or ACC. Identification results were concordant for 29 of 33 isolates (87.88%). Discordant results included a *Klebsiella variicola* pathogen that was erroneously identified as *Enterobacter* spp, and three *Escherichia coli* organisms that were not identified by ACC. Antimicrobial susceptibility testing data were available for a total of 28 of 33 samples (84.85%). A total of 434 microorganism-antimicrobial combinations were analyzed. Results were as follows: CA 97.93% (425/434), VME 0.23% (1/434), ME 1.15% (5/434), and mE 0.69% (3/434) (*Table 3*). No discrepancies were observed with respect to resistance to carbapenems.

Real and hypothetical impact on clinical management of patients

Impact on clinical management of patients of ACC was evaluated by comparing ACC to the standard method for TTR, duration of empiric antimicrobial therapy, and switch to targeted therapy. A total of 28 of 33 patients, for which ACC AST results were available, were evaluated. It was found that TTR (mean ± SD) of AST results obtained by ACC (27.14±6.90 h) was significantly shorter ($p<0.001$, Δ = 19.96 h, 95% CI: 24.71-15.22) than that obtained by standard method (47.10±11.92 h). It is worth noting that the standard deviation was also reduced in ACC when compared to standard of care (6.97 vs 11.92 h).

Empiric therapy included piperacillin/tazobactam in 11 patients and carbapenems in another 11 cases. Three patients were treated with quinolones, 2 with ceftriaxone, and one with tigecycline plus amikacin.

A switch from empiric to targeted therapy was observed in 14 patients (50.00%): de-escalation in 8 patients and escalation in 6. In these patients, duration of empiric therapy was 37.73±19.87 h.

After a mean time of 18.88 h from ACC final report, piperacillin/tazobactam was switched to 3rd cephalosporins in 2/11 patients, saving 5.45 PDD compared to the same switch after standard AST report. De-escalation of carbapenems was observed in 3/11 patients, after a mean time of 10.56 h from ACC results, with 5.28 PDD saved. In these cases, errors by ACC were as follow: colistin MEs in 2 *E. coli* isolates, in one case ertapenem was switched to ceftriaxone, and in the other ceftriaxone was switched to meropenem; gentamycin ME and aztreonam mE in a *P. aeruginosa* isolate in a patient in which piperacillin/tazobactam was switched to ceftolozane/tazobactam. Thus, in these cases, errors observed did not negatively impact the final therapeutic decision.

Piperacillin/tazobactam was maintained in 9/11 patients and carbapenems in 8/11. Based on a review of patients' charts, a switch to targeted therapy with a narrow-spectrum antibiotic would have been theoretically feasible in 7/9 cases treated with piperacillin/tazobactam and in 4/8 patients treated with carbapenems.

Table 4 shows the hypothetical sparing of PDD obtainable by ACC, compared to the standard method. This was assessed assuming that, both in patients in whom the de-escalation was done (2 treated with piperacillin/tazobactam and 3 treated with carbapenems) and in those in whom it was not done (7 patients treated with piperacillin/tazobactam and 4 patients treated with carbapenems), empiric therapy would have been switched to targeted therapy

Table 3 - *Very major (VME), major (ME), and minor (mE) errors observed among 434 microorganism-antimicrobial combinations when comparing results from the same bacterial isolate using ACC and standard method.*

Isolate	*VME*	*ME*	*mE*
Escherichia coli	TZP	-	-
Escherichia coli	-	Colistin	-
Escherichia coli	-	Colistin	-
Escherichia coli	-	Ciprofloxacin	-
Escherichia coli	-	-	Cefepime and Ceftazidime
Enterobacter spp	-	Colistin	-
Pseudomonas aeruginosa	-	Gentamycin	Aztreonam
Total	1 (0.23%)	5 (1.15%)	3 (0.69%)

TZP, piperacillin/tazobactam.
-, none.

Table 4 - *Hypothetical broad-spectrum antibiotic prescribed daily doses (PDD) sparing in patients receiving empirical therapy of piperacillin/tazobactam and carbapenem.*

Empirical treatment	*Patient*	*ACC Report (hours)*	*Standard Report (hours)*	*Time difference (hours)*	*PDD saved/time difference*
Piperacillin/tazobactam					
	1	14.40	44.15	29.75	3.72
	2	26.00	40.07	14.07	1.76
	3	37.55	52.52	14.97	1.87
	4	18.55	73.02	54.47	6.81
	5	25.77	41.03	15.26	1.91
	6[a]	25.25	61.50	36.55	4.57
	7	29.38	44.32	14.94	1.87
	8	26.83	38.50	11.67	1.46
	9[a]	24.45	54.23	29.78	3.72
Total	9	25.35 ± 6.47[b]	49.93 ± 11.53[b]	24.57 ± 14.29[b]	*27.69*
Carbapenem[c]					
	1	24.85	38.58	13.73	1.72
	2	25.53	87.82	62.29	7.79
	3	24.45	54.23	29.78	3.72
	4[a]	28.03	44.37	16.34	2.04
	5[a]	37.55	52.52	14.97	1.87
	6[a,]	16.15	48.98	32.83	1.37
	7	42.30	56.08	13.78	0.57
Total	7	28.41 ± 8.79[b]	54.65 ± 15.83[b]	26.25 ± 17.76[b]	*19.08*

[a]Patient in whom de-escalation was actually done, although not immediately after ACC report.
[b]Mean ± SD.
[c]All patients were treated with meropenem, except patients 6 and 7, receiving ertapenem.

soon after the ACC final report. It was found that, based on ACC results, 27.69 PDD of piperacillin/tazobactam and 19.08 PDD of carbapenems could had been spared.

DISCUSSION

Early effective therapy is critical to a successful outcome in septic patients (Kumar *et al.*, 2006; Emonet & Schrenzel, 2011), and clinicians are forced into antibiotic broad-spectrum therapy before etiology becomes available, highlighting the need for rapid ID and AST results.
Recently, ACC has been approved to speed up positive BC workflow and its accuracy and usefulness have been described (Pancholi *et al.*, 2018; Lutgring *et al.*, 2018; Marschal *et al.*, 2017). It has been hypothesized that ACC could have a positive impact on the time to effective and definitive antimicrobial therapy in bloodstream infections from resistant Gram-negative bacteria (Henig *et al.*, 2018), although its impact is likely lessened by the availability of other rapid diagnostic tests (Henig *et al.*, 2019).
Italy is regrettably characterized by a high prevalence of multi-drug resistant organisms like methicillin-resistant *S. aureus* (Annual Report EARS-NET, 2018; Campanile *et al.*, 2015), vancomycin-resistant *Enterococcus* spp. (Annual Report EARS-NET, 2018), and carbapenemase-producing enterobacteria (CPE) (Grundmann *et al.*, 2017; Giani *et al.*, 2017), mainly *K. pneumoniae* (Giani *et al.*, 2017). In BC positive for these isolates, rapid molecular tests for detection of resistance genes are of crucial importance to speed up appropriate therapy, so that phenotypic AST results can be delivered according to standard laboratory workflow. In fact, although Pantel *et al*. have shown that ACC can be useful for resistance detection

(Pantel *et al*., 2018), the ACC antibiotics panel available until 2019 did not provide information on new drugs used in clinical practice for the treatment of infections caused by multi-resistant organisms. The recent implementation of an ACC panel with tigecycline, ceftazidime/avibactam and ceftolozane/tazobactam will be useful to extend the workflow to these microorganisms as well, in a setting where these pathogens are frequently found.

On the contrary, in case of Gram-negative pathogens different from CPE, AST is essential for switching from empiric to targeted therapy, provided there is close collaboration with clinicians for an immediate de-escalation of antimicrobial therapy. Thus, considering our epidemiology, the panel of rapid tests available in the laboratory, the antimicrobials included in the ACC panel, and the costs of this technology, it makes sense to perform the test only in cases in which clinical management of patients could really benefit from ACC results. In the study period, according to the strict workflow applied to select samples to be tested (*Figure 1*), ACC could have been employed for a total of 48 patients. Unfortunately, in 15 patients, the ACC test was not performed due to our limited operating hours, emphasizing that to really take advantage of this test, and of any rapid test, a laboratory should operate 24/7. However, it is worth mentioning that a statistical significance in TTR was also achieved with a small sample size of 33 patients.

In this study 87.88% of pathogens were correctly identified and high CA of 97.93% was observed, in line with previous studies (Charnot-Katsikas *et al*., 2018; Pancholi *et al*., 2018; Marschal *et al*., 2017). Indeed, misidentification between *K. variicola* and *Enterobacter* species has been described in other studies, when comparing rapid molecular tests with standard biochemical identification (Ledeboer *et al*., 2015). Interestingly, AST results reported by ACC for the misidentified *K. variicola* isolate were fully correct. In 3 cases no ID was obtained by ACC, while AST failed in 5/33 cases, possibly due to the presence of antimicrobials, or absence or excessive bacterial growth, as suggested by the analysis of the bacterial growth curves (data not shown). Descours *et al.* suggested that improvements in AST algorithms are needed to implement this system in the routine workflow (Descours *et al*., 2018). The high CA (97.93%) resulted in a few errors, but these errors did not negatively impact the final therapeutic decision. The fact that the majority of ME were observed for colistin supports avoiding the use of ACC in the case of CPE pathogens, in which colistin represents a therapeutic option. The only VME was related to piperacillin/tazobactam for one *E. coli* isolate, resistant to 3rd cephalosporins, producing extended-beta-lactamase, in which empiric treatment with meropenem was correctly maintained after ACC and standard AST results.

The results of the present study demonstrate that ACC significantly reduced TTR by approximately one day with respect to the standard method, as previously demonstrated by Charnot-Katsikas *et al.* (Charnot-Katsikas *et al*., 2018). On average, hospital microbiology laboratories take approximately one day from the time of sample collection to obtain a Gram stain, two days for ID, and three days for AST results. Our laboratory has a faster workflow thanks to a laboratory automation system (De Socio *et al*., 2018). Nevertheless, we found that the anticipation of AST results by ACC could impact on the sparing of broad-spectrum antimicrobial agents provided that a switch to a targeted therapy occurs soon after the ACC report. In fact, in our study population the duration of empiric therapy was as short as 37.73 h. This time is inferior to the TTR of 47.10 h found in this study for the standard protocol, and shorter than the mean time of empiric therapy of 54.8 h found in a previous study performed on 100 patients in the same hospital (De Socio *et al*., 2018).

It has been demonstrated that empiric antibiotic therapy is frequently inappropriate, and this leads to an increased risk of mortality, prolonged hospitalizations, and incremental costs (Diamantis *et al*., 2012; Paul *et al*., 2010). In addition, the prolonged use of broad-spectrum antibiotics is a known risk factor for the development and spread of antimicrobial resistance (Ventola, 2015; Zaman *et al*., 2017). We found that, in 5 cases in which empiric therapy with broad-spectrum antibiotics was promptly de-escalated, 5.45 PDD of piperacillin/tazobactam and 5.28 PDD of carbapenems had been spared due to the rapid AST results delivered by ACC. Analyses of cases in which antibiotics were continued, after either ACC or standard AST report, showed that spared PPD values could have been significantly higher if therapeutic management of patients had been done properly (García-Rodríguez *et al*., 2019). In fact, in 11 cases therapy was not changed, despite ACC results. The reason could be found in the observational nature of the study, performed in a hospital lacking a defined AS program, and without a specific therapeutic protocol based on ACC results. Thus, therapeutic decisions were made by clinicians according to their different ward practices, and not according to a shared therapeutic algorithm. This issue highlights the crucial importance and need of AS programs in hospitals, and of a sepsis team translating laboratory results into rapid and effective intervention for septic patients (Humphries *et al*., 2019). Specifically, ACC can assist clinicians in optimizing antibiotic use with targeted narrow-spectrum antimicrobial therapy in a very short time (just over a day).

There are some limitations in this study. First, the limited sample size, discussed above. Second, only patients with monomicrobial infections were included in the workflow, while the test can also be employed in selected polymicrobial sepsis. Finally, the study was not interventional and there was no specific agreement on therapeutic decisions, given the lack of defined hospital AS programs.

In conclusion, this study demonstrates that even in laboratories where automation and other rapid diagnostic equipment are available, ACC, included in a specific workflow to process positive BC, could play an important role in reducing the time for switching from empiric to targeted therapy. This could have a crucial impact on broad-spectrum antibiotic sparing, provided there is 24/7 laboratory operating time, strict collaboration between clinical microbiologists and clinicians, and implementation of an effective hospital AS program.

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The authors declare that they have no conflicts of interest.

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