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Original Article

The real-world impact of the BioFire FilmArray blood culture identification 2 panel on antimicrobial stewardship among patients with bloodstream infections in intensive care units with a high burden of drug-resistant pathogens

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KEYWORDS
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Abstract *Background:* The increasing prevalence of drug-resistant pathogens leads to delays in adequate antimicrobial treatment in intensive care units (ICU). The real-world influence of the BioFire FilmArray Blood Culture Identification 2 (BCID2) panel on pathogen identification, diagnostic concordance with conventional culture methods, and antimicrobial stewardship in the ICU remains unexplored.

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Critically ill;
Drug-resistant pathogen;
Intensive care units;
Multiplex polymerase chain reactio

Methods: This retrospective observational study, conducted from July 2021 to August 2023, involved adult ICU patients with positive blood cultures who underwent BCID2 testing. The concordance between BCID2 and conventional culture results was examined, and its impact on antimicrobial stewardship was assessed through a comprehensive retrospective review of patient records by intensivists.

Results: A total of 129 blood specimens from 113 patients were analysed. Among these patients, a high proportion of drug-resistant strains were noted, including carbapenem-resistant *Klebsiella pneumoniae* (CRKP) (57.1%), carbapenem-resistant *Acinetobacter calcoaceticus-baumannii* complex (100%), methicillin-resistant *Staphylococcus aureus* (MRSA) (70%), and vancomycin-resistant *Enterococcus faecium* (VRE) (100%). The time from blood culture collection to obtaining BCID2 results was significantly shorter than conventional culture (46.2 h vs. 86.9 h, $p < 0.001$). BCID2 demonstrated 100% concordance in genotype—phenotype correlation in antimicrobial resistance (AMR) for CRKP, carbapenem-resistant *Escherichia coli*, MRSA, and VRE. A total of 40.5% of patients received inadequate empirical antimicrobial treatment. The antimicrobial regimen was adjusted or confirmed in 55.4% of patients following the BCID2 results.

Conclusions: In the context of a high burden of drug-resistant pathogens, BCID2 demonstrated rapid pathogen and AMR detection, with a noticeable impact on antimicrobial stewardship in BSI in the ICU.

Introduction

Bloodstream infection (BSI) in critically ill patients require timely diagnosis and antibiotic treatment to effectively improve survival.[1,2] However, an escalating antimicrobial resistance in the intensive care unit (ICU) has been noted in recent years.[3,4] The "ESKAPE" pathogens (*Enterococcus faecium, Staphylococcus aureus, Klebsiella pneumoniae, Acinetobacter baumannii, Pseudomonas aeruginosa*, and Enterobacter species) continue to be leading causes of multidrug-resistant organisms (MDROs) infections worldwide.[5,6]

Antimicrobial resistance is related to delays in achieving appropriate antimicrobial therapy and heightened mortality.[6,7] In conjunction with the high disease severity and mortality in ICU patients with BSI, antimicrobial resistance leads to a substantial rise in the use of broad-spectrum antibiotics, exacerbating the issue by promoting the selection of antimicrobial-resistant pathogens.[4] Thus, early identification of bacterial strains and the use of appropriate antibiotics are essential.

Conventionally, bacterial pathogens are identified through blood cultures, a time-consuming process involving multiple incubation steps for pathogen identification and antimicrobial susceptibility testing (AST). Multiplex PCR (mPCR) assays have been developed to rapidly identify pathogens in flagged blood cultures, eliminating the need for bacterial subculture and facilitating the prompt and precise identification of pathogens.[8,9] The BioFire FilmArray Blood Culture Identification 2 (BCID2) panel (BioFire Diagnostics, bioMérieux, Salt Lake City, Utah) is a second-generation mPCR system capable of detecting 43 targets, including 26 bacteria, 7 yeast, and 10 antibiotic resistance genes. The system provides results within 1 h for positively flagged blood cultures.[9] Supplementary Table 1 illustrates the differences between its first version, the BCID, and the new version, BCID2 panels. Previous literature had reported the performance and benefit of BCID2 in clinical care,[10—12] with the ability to detect the most common 'ESKAPE' pathogens in the ICU and more antibiotic resistance genes than BCID. However, limited studies have focused on the application of BCID2 in critically ill patients with BSI in the ICU and its role in antimicrobial stewardship. This study aims to evaluate the real-world impact of BCID2 on patients with BSI in the ICU.

Materials and methods

Patients and study design

This retrospective observational study was conducted at a tertiary medical center in central Taiwan, China Medical University Hospital (CMUH). Critically ill adult patients (aged ≥18) diagnosed with BSI who underwent BCID2 Panel from July 2021 to August 2023 were enrolled. During the study period, all ICU patients with positive blood cultures underwent BCID2 testing. The flagged blood culture was tested with BCID2 after obtaining informed consent from the patients or their family. Flagged blood culture samples with suspect contamination were excluded.

This study received approval from the Institutional Review Board (IRB) of the CMUH (CMUH108-REC2-111, CMUH111-REC1-074). It was conducted in accordance with the Declaration of Helsinki and followed the Strengthening the Reporting of Observational studies in Epidemiology (STROBE) guidelines.

Definitions and data collection

Blood culture collection was initiated promptly when patients exhibiting systemic signs of infection, and BSI was defined as positive blood culture.[13] In this study,

Table 1 Demographic data of patients with bacteraemia undergoing the BioFire FilmArray® Blood Culture Identification Panel 2 (BCID2) for pathogen identification in the ICU.

Characteristic	n = 113
Age (years), median (Q1, Q3)	68.5 (60.0, 78.1)
Sex, male, *n* (%)	82 (72.6)
BMI (kg/m^2), median (Q1, Q3)	23.6 (20.0, 26.9)
Modified Charlson score, median (Q1, Q3)	6 (4, 8)
Comorbidities, *n* (%)	
Hypertension	53 (46.9)
Diabetes mellitus	53 (46.9)
Immunocompromised [a]	42 (37.2)
Solid cancer	29 (25.7)
Hematologic malignancy	13 (11.5)
Chronic kidney disease	28 (24.8)
Coronary artery disease/Congestive heart failure	17 (15.0)
Liver cirrhosis	14 (12.4)
Chronic obstructive pulmonary disease	11 (9.7)
Cerebral vascular accident	8 (7.1)
Risk factors, *n* (%)	
History of hospital admission within 90 days	42 (37.2)
Chronic dialysis	35 (31.0)
Intravenous antibiotic(s) exposure within 90 days	31 (27.4)
History of operation within 90 days	21 (18.6)
History of ICU admission within 90 days	8 (7.1)
Nursing home/RCW resident	6 (5.3)
Any one of above risk factors	69 (61.1)
Parenteral nutrition, *n* (%)	21 (18.6)
Chemotherapy, *n* (%)	10 (8.8)
Infection focus, *n* (%)	
Intraabdominal infection	38 (33.6)
Pneumonia	34 (30.1)
Urinary tract infection	13 (11.5)
Catheter related	9 (8.0)
Soft tissue infection	2 (1.8)
Multiple focus	6 (5.3)
Unknown	11 (9.7)
Disease severity on bacteraemia index date	
APACHE II score, median (Q1, Q3)	29 (23, 32)
SOFA score, median (Q1, Q3)	13 (9, 15)
Septic shock, *n* (%)	82 (72.6)
Inflammatory markers	
CRP (mg/L), median (Q1, Q3)	16.0 (5.5, 23.9)
Procalcitonin (ng/mL), median (Q1, Q3) (*n* = 96)	24.9 (5.7, 75.6)
Organ support, *n* (%)	
Invasive mechanical ventilation	107 (94.7%)
Continuous renal replacement therapy	20 (17.7)
Outcome	
ICU LOS (*n* = 61) [b]	13 (8—22.5)
ICU mortality, *n* (%)	52 (46.0)
Hospital LOS (*n* = 57) [b]	36 (18—76)
Hospital mortality, *n* (%)	56 (49.6)

[a] Hematologic malignancies, Chronic steroid use (prednisolone 5 mg/day or equivalent >1 month or >30 mg/day) or other immunosuppressive therapy for diseases such as connective tissue disease, rheumatic disease, or solid organ transplantation.

[b] The assessment of ICU and hospital stays did not include patients who died during hospitalization.

APACHE II, Acute Physiology and Chronic Health Evaluation II; BMI, body mass index; CRP, C-reactive protein; ICU, intensive care unit; LOS, length of stay; RCW, respiratory care ward; SOFA, Sequential Organ Failure Assessment.

contamination was defined as follows: if only one set of at least two sets of blood cultures was positive for coagulase-negative staphylococci (CoNS), or if CoNS was detected in a single set of blood culture.[14]

Patient characteristics, the primary infection focus of BSI, risk factors for acquiring drug-resistant pathogens including antibiotic exposure history and recent hospital admission, results of BCID2 and the standard of care blood culture, antibiotic regimen, disease severity, presence of organ failure and level of organ support were retrospectively collected from electronic medical records. Disease severity was assessed with Acute Physiologic Assessment and Chronic Health Evaluation II (APACHE II) score and Sequential Organ Failure Assessment (SOFA) Score on the BSI index date.

Microbiological testing

The blood culture bottles were placed in a BD BACTEC™ FX Blood Culture System (Becton Dickinson, Sparks, MD, USA). When the BACTEC FX system flags a blood culture as positive, indicating microbial growth, a Gram stain is conducted to verify the presence of microorganisms including Gram-positive (GP) or Gram-negative (GN) bacteria. In addition to the conventional culture methods, positive blood culture bottles were analysed using the BCID2 panel in the enrolled patients.

The BCID2 procedure adhered to the manufacturer's guidelines and was executed at the clinical microbiology laboratory of CMUH. Positive bacterial and genetic markers detection results were reported in a qualitative manner.

In conventional methods, microbial identification to the species level was carried out using matrix-assisted laser desorption ionization-time of flight mass spectrometry (MALDI-TOF MS) Biotyper system (Bruker Microflex LT/SH, Bruker Daltonics GmbH) with colonies, and antimicrobial susceptibility testing was conducted using a commercial susceptibility system (BD Phoenix M50 Automated Microbiology System, Becton Dickinson, Sparks, MD, USA).

Performance measures of BCID2

The bacterial analyte culture findings, serving as the standard of care, were employed as the gold standard and reference method for bacterial detection in the analysis of the performance of BCID2. A BCID2 result was categorized as a true positive or true negative when it aligned with the standard-of-care investigation results. A BCID2 result is deemed false positive if it is positive while the culture result is negative. Conversely, a BCID2 result is deemed false negative if it is negative while the culture result is positive. Concordance was established when the BCID2 results for bacterial detection aligned with the results of the conventional culture.

Clinical impact of BCID2 on antimicrobial therapy

During the study period, the attending intensivist administered empiric antibiotic therapy for individuals presenting with BSI in ICU. Antibiotics were adjusted based on the results obtained from BCID2 and final conventional culture results.

Three distinct intensivists conducted a retrospective evaluation of the BCID2 results' influence on antimicrobial treatment by reviewing patient medical records. When discrepancies arose between two reviewers, a third intensivist was consulted to reach a consensus. "Adequate antimicrobial therapy" was characterized by the administration of at least one antibiotic deemed appropriate according to the final AST. The term "adequate but could be de-escalated" refers to scenarios where treatment could potentially shift to antibiotics with a narrower spectrum.

Statistical analysis

All statistical analyses were conducted using SPSS ver. 25 (SPSS Inc., Chicago, IL, USA). Categorical variables were presented as numbers and percentages. Continuous data were presented as the median and interquartile range, and

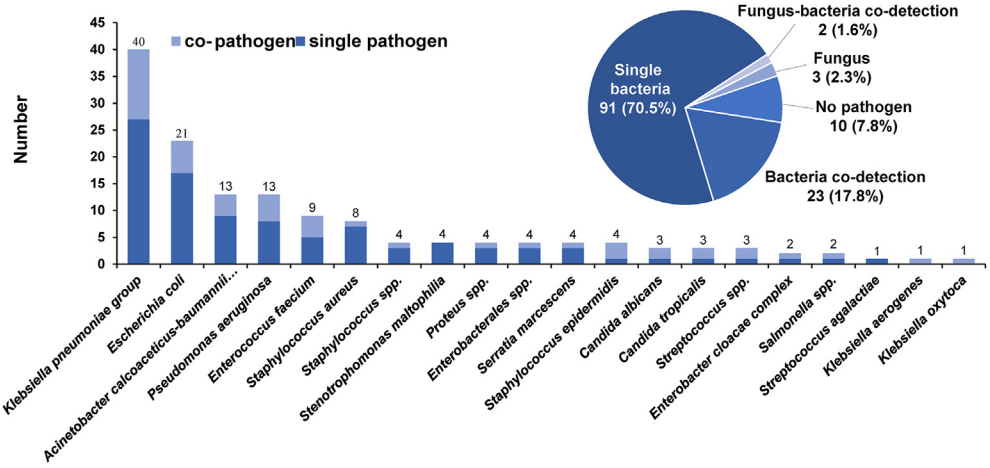

Fig. 1. Pathogens detected by BioFire FilmArray® Blood Culture Identification Panel 2 (BCID2) panel among 129 blood specimens obtained from 113 patients with bloodstream infection. A single pathogen is indicated in deep blue; co-pathogens are indicated in a lighter colour.

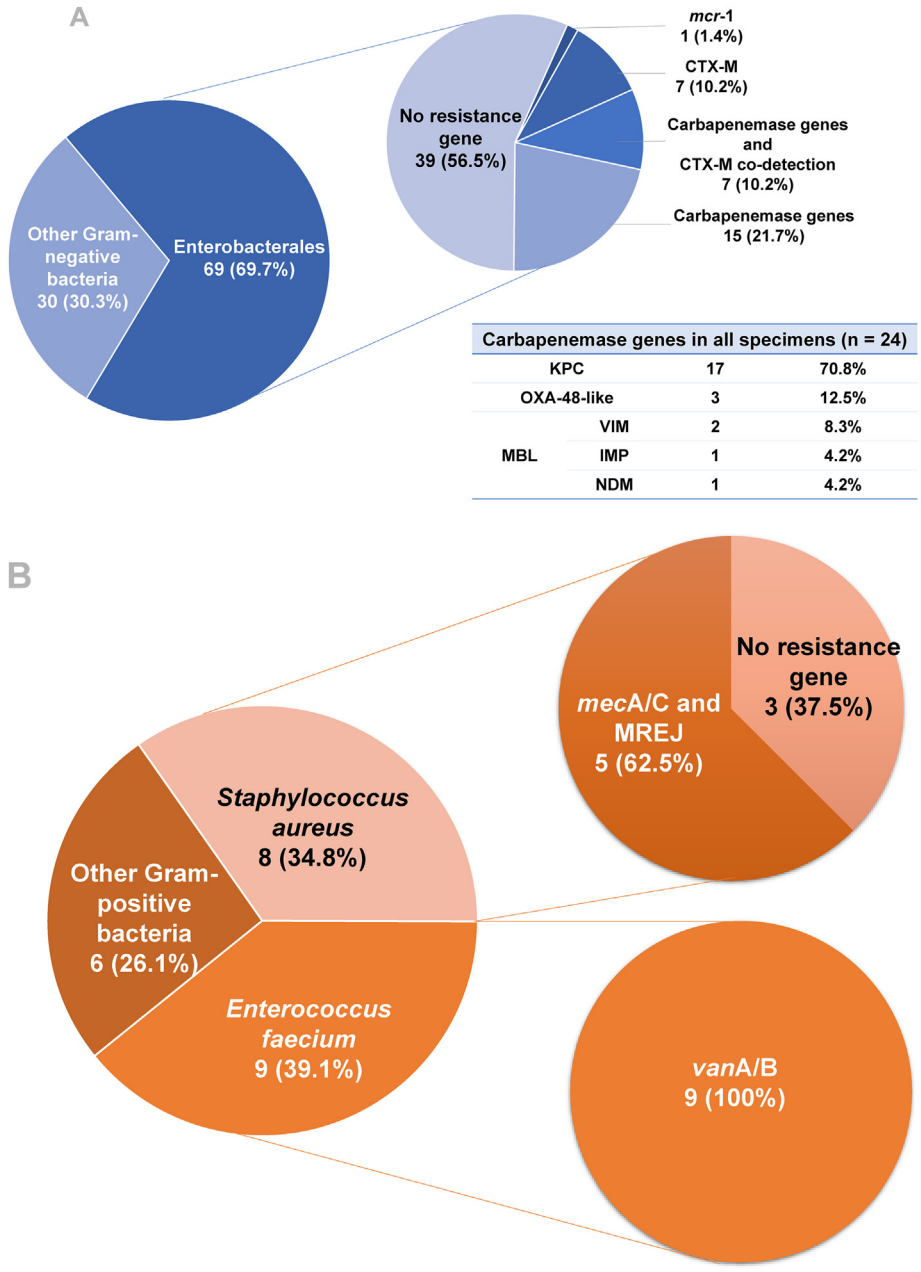

Fig. 2. (A) Distribution and analysis of total detected antimicrobial resistance genes by BioFire FilmArray® Blood Culture Identification Panel 2 (BCID2) panel among 99 specimens from patients with Gram-negative bacteraemia, with a focus on Enterobacterales. And distribution analysis of total carbapenemase genes in all specimens (B) Distribution and analysis of total detected antimicrobial resistance genes by BCID2 panel among 23 specimens from patients with Gram-positive bacteraemia, with a focus on *Staphylococcus aureus* and *Enterococcus faecium*. MBL, metallo-β-lactamase.

group differences were assessed using the Mann—Whitney U test. All tests were two-tailed, and statistical significance was indicated by a P value < 0.05.

Results

Demographic characteristics

This study analysed a total of 129 blood specimens obtained from 113 patients with BSI. Among these patients, 82 were male (72.6%), with a median age of 68.5 years (Q1, Q3 60.0, 78.1). The most prevalent comorbidities included hypertension (46.9%), diabetes mellitus (46.9%), immunocompromised status (37.2%), and solid cancer (25.7%). Notably, 37.2% of patients had a history of recent hospital admission, and 27.4% had been exposed to intravenous antibiotics within the past 90 days. Of the total, 69 (61.1%) patients displayed at least one risk factor for acquiring drug-resistant pathogens (Table 1).

Intra-abdominal infection (33.6%) and pneumonia (30.1%) were the most common infection focuses. The

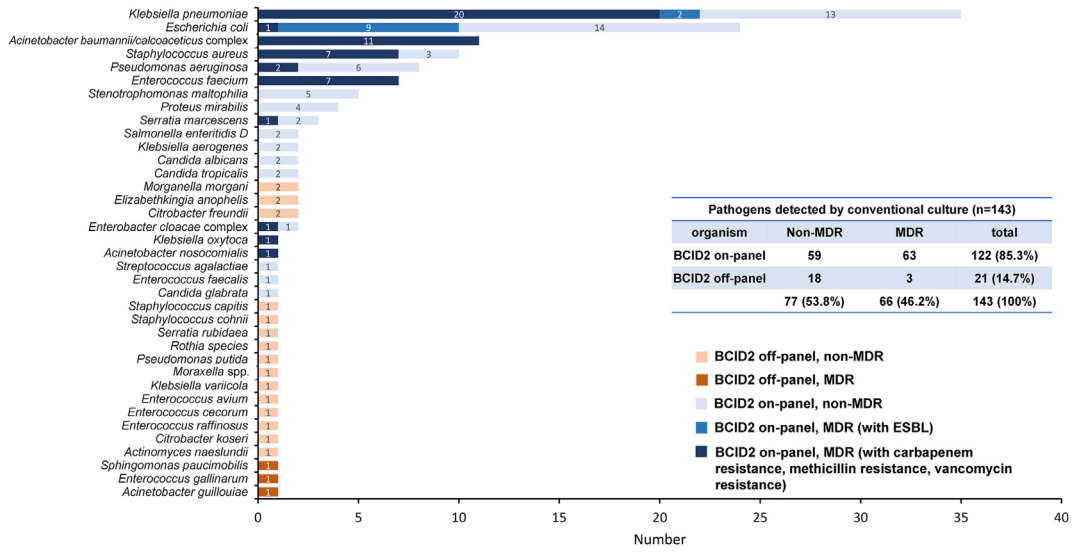

Fig. 3. Pathogens detected by conventional culture and antimicrobial susceptibility testing. Labelled different colors whether the pathogens detectable by the BioFire FilmArray® Blood Culture Identification Panel 2 (BCID2) panel and antimicrobial resistance (Carbapenem/Methicillin/Vancomycin -resistance) and extended-spectrum β-lactamase are present or not. The blue color indicates detectability by the BCID2 panel, while the brown color indicates non-detectability by the BCID2 panel. Additionally, darker colors represent drug resistance, while lighter colors indicate the absence of drug resistance. ESBL, extended spectrum beta lactamase, MDR, multiple drug resistance.

median C-reactive protein level was 16.0 (Q1, Q3 5.5, 23.9), and the median procalcitonin level was 24.9 (Q1, Q3 5.7, 75.6). The median APACHE II score on the BSI index date was 29 (Q1, Q3 23, 32), and the SOFA score was 13 (Q1, Q3 9, 15). Septic shock developed in 82 (72.6%) patients. Invasive mechanical ventilation was required for 107 (94.7%) patients, and 20 (17.7%) underwent continuous renal replacement therapy (Table 1).

The median ICU length of stay (LOS) after BSI index date was 13 days (Q1, Q3 8, 22.5), and the hospital LOS was 36 days (Q1, Q3 18, 76). The ICU and hospital mortality rates were 46.0% and 49.6%, respectively.

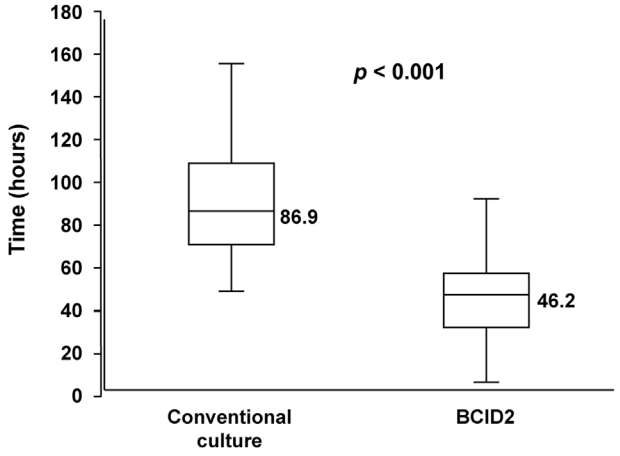

Fig. 4. Time from sampling to diagnostic results by BioFire FilmArray® Blood Culture Identification Panel 2 panel (BCID2) and conventional culture.

Microbiological outcomes

BCID2 findings

Among all blood culture specimens, BCID2 yielded positive results in 119 (92.2%) specimens, with majority caused by single bacteria (70.5%). The most frequently identified GN bacteria included the *K. pneumoniae* group (n = 40), *Escherichia coli* (n = 21), *Acinetobacter calcoaceticus-baumannii* complex (n = 13), and *P. aeruginosa* (n = 13) (Fig. 1). Antimicrobial resistance genes were identified in 47 (36.4%) specimens. Among these, 21.3% (10/47) exhibited more than two resistance genes (Fig. S1). The most detected gene was bla_{KPC} (n = 17, 28.8%), followed by bla_{CTX-M} (n = 15, 25.4%) and *van*A/B (n = 9, 15.3%). metallo-β-lactamases (MBLs, bla_{NDM}, bla_{IMP}, bla_{VIM}) accounted for 6.8% (n = 4) of the detected resistance genes (Fig. S1). We further analysed the resistance genes in different bacterial strains. The most common GN bacterial pathogens were Enterobacterales (n = 69, 69.7%). Among these, carbapenemase genes were detected in 21.7%, bla_{CTX-M} were found in 10.2%, and co-detection of carbapenemase genes and bla_{CTX-M} was observed in 10.2% (Fig. 2A). *E. faecium* (n = 9) and *S. aureus* (n = 8) were the most common GP bacteria. All *E. faecium* carried *van*A/B gene (n = 9, 100%) and 62.5% of *S. aureus* carried *mec*A/C and MREJ genes (n = 5, 62.5%) (Fig. 2B).

Conventional culture findings

Among the 143 pathogens detected by conventional culture, drug-resistant pathogens account for 46.2% (n = 66). The most frequently detected GN bacteria were *K. pneumoniae* (n = 35), *E. coli* (n = 24), *A. calcoaceticus-baumannii* complex (n = 11), and *P. aeruginosa* (n = 8). The

Table 2 Performance of the BCID2 Panel for the identification of microorganisms versus the conventional culture as gold standard. Sensitivity was calculated as [true positive/(true positive + false negative)] X 100%, while specificity was calculated as [true negative/(true negative + false positive)] X 100%. The positive predictive value (PPV) was calculated as [true positive/(true positive + false positive)] X 100%, and the negative predictive value (NPV) was calculated as [true negative/(true negative + false negative)] X 100%. The concordance rate (%) was calculated as the number of concordant results divided by the number of concordant and discordant results [total number of tests, $n = 129$]. BCID2, the BioFire FilmArray® Blood Culture Identification Panel 2.

	Organisms	True positive (Culture +/BCID2 +)	False positive (Culture −/BCID2 +)	False negative (Culture +/BCID2 -)	True negative (Culture -/BCID2 -)	Se (%)	Sp (%)	PPV (%)	NPV (%)	Concordance rate, % (no. of concordant results)
Gram-positive bacteria	*Staphylococcus aureus*	8	0	2	119	80	100	100	98	98.4 (127)
	Staphylococcus epidermidis	1	3	0	125	100	98	25	100	97.7 (126)
	Staphylococcus lugdunensis	0	0	0	129	—	100	—	100	100 (129)
	Streptococcus agalactiae	1	1	0	127	100	99	50	100	99.2 (128)
	Streptococcus pneumoniae	0	0	0	129	—	100	—	100	100 (129)
	Streptococcus pyogenes	0	0	0	129	—	100	—	100	100 (129)
	Enterococcus faecalis	1	0	0	128	100	100	100	100	100 (129)
	Enterococcus faecium	6	3	1	119	86	98	67	99	96.9 (125)
	Listeria monocytogenes	0	0	0	129	—	100	—	100	100 (129)
Enterobacterales	*Enterobacter cloacae* complex	2	0	0	127	100	100	100	100	100 (129)
	Escherichia coli	21	2	2	104	91	98	91	98	96.9 (125)
	Klebsiella aerogenes	1	0	1	127	50	100	100	99	99.2 (128)
	Klebsiella oxytoca	1	0	0	128	100	100	100	100	100 (129)
	Klebsiella pneumoniae group	34	6	0	89	100	94	85	100	95.3 (123)
	Proteus spp.	4	0	0	125	100	100	100	100	100 (129)
	Salmonella spp.	2	0	0	127	100	100	100	100	100 (129)
	Serratia marcescens	3	1	0	125	100	99	75	100	99.2 (128)
Glucose nonfermenting gram-negative bacteria	*Acinetobacter calcoaceticus-baumannii* complex	11	2	1	115	92	98	85	99	97.7 (126)

	Pseudomonas aeruginosa	7	6	2	114	78	95	54	98	93.8 (121)
Other	*Bacteroides fragilis*	0	0	0	129	—	100	—	100	100 (129)
	Haemophilus influenzae	0	0	0	129	—	100	—	100	100 (129)
	Neisseria meningitidis	0	0	0	129	—	100	—	100	100 (129)
	Stenotrophomonas maltophilia	4	0	1	124	80	100	100	99	99.2 (128)
Fungus	*Candida albicans*	2	1	0	126	100	99	67	100	99.2 (128)
	Candida auris	0	0	0	129	—	100	—	100	100 (129)
	Candida glabrata	1	0	0	128	100	100	100	100	100 (129)
	Candida krusei	0	0	0	129	—	100	—	100	100 (129)
	Candida parapsilosis	0	0	0	129	—	100	—	100	100 (129)
	Candida tropicalis	2	1	0	126	100	99	67	100	99.2 (128)
	Cryptococcus	0	0	0	129	—	100	—	100	100 (129)

Table 3 Correlation of antimicrobial resistance genotype (detected by BioFire FilmArray® Blood Culture Identification Panel 2) and phenotype (detected by conventional culture with antimicrobial susceptibility testing) in pathogens detected by both methods.

Pathogens		Concordance rate (%)			
		ESBL	CRE	MRSA	VRE
Enterobacterales	*Enterobacter cloacae* complex	100	66.7	–	–
	Escherichia coli	85	100[a]	–	–
	Klebsiella aerogenes	100	100	–	–
	Klebsiella oxytoca	–	100[b]	–	–
	Klebsiella pneumoniae group	71.4	100[c]	–	–
	Proteus spp.	100	100	–	–
	Serratia marcescens	100	100[d]	–	–
Gram-positive bacteria	*Staphylococcus aureus*	–	–	100[e]	–
	Enterococcus faecium	–	–	–	100[f]

[a] KPC ($n = 1$).
[b] VIM ($n = 1$).
[c] KPC ($n = 16$), OXA-48-like ($n = 3$), IMP ($n = 1$), NDM ($n = 1$).
[d] VIM (1).
[e] *mec*A/C and MREJ ($n = 4$), *mec*A/C ($n = 2$).
[f] *van*A/B ($n = 6$).
BCID2, BioFire FilmArray® Blood Culture Identification Panel 2; CRE, Carbapenem-resistant Enterobacterales; ESBL, Extended Spectrum Beta Lactamase; MRSA, Methicillin-resistant *Staphylococcus aureus*; VRE, Vancomycin-resistant *Enterococcus*.
Resistance genes detected by BCID2 Panel.

most common GP pathogens were *S. aureus* (n = 10) and *E. faecium* (n = 7) (Fig. 3). Among the common pathogens detected in the ICU, 20 out of 35 (57.1%) *K. pneumoniae* were carbapenem resistant, and 2 (5.7%) were classified as extended spectrum beta lactamase (ESBL) strains. For *E. coli*, 9 out of 24 (37.5%) were ESBL, and 1 (4.2%) exhibited carbapenem resistance. All detected *A. calcoaceticus-baumannii* complex were carbapenem resistant strains. Regarding *P. aeruginosa*, 2 out of 8 (25%) were carbapenem resistant. Seven out of 10 (70%) *S. aureus* exhibited oxacillin resistance, and all *E. faecium* were vancomycin resistant. Moreover, 14.7% (21/143) of off-panel organisms, which the BCID2 Panel is not designed to detect, were identified by conventional culture, with the most frequently detected ones being *Elizabethkingia anophelis* (n = 2), *Morganella morgani* (n = 2), and *Citrobacter freundii* (n = 2) (Fig. 3).

Performance of BCID2

The time from sample collection to a complete result report was 46.2 h (Q1, Q3 30.4, 56.7) and 86.9 h (Q1, Q3 70.6, 110.2) for BCID2 and conventional culture with complete AST (Fig. 4).

Using the results of conventional blood culture as the gold standard for comparison, the specificity and NPV of BCID2 were both higher than 90% for all species (Table 2). Among common pathogens in the ICU, the sensitivity is greater than 90% in the *K. pneumoniae* group, *E. coli*, and *A. calcoaceticus-baumannii* complex. As for *P. aeruginosa*, *E.faecium*, and *S. aureus*, their sensitivities are 78%, 86%, and 80%, respectively.

The correlation between the antimicrobial resistance genotype, as detected by BCID2, and the phenotype, as detected by conventional culture with AST, is shown in Table 3. Among the common pathogens in the ICU with detectable resistance genes, the concordance rates are 71.4% and 85% for ESBL *K. pneumoniae* and ESBL *E. Coli*. For CRKP, carbapenem-resistant *E. Coli*, MRSA, and VRE, the concordance rates are 100%. Detailed information on genotype—phenotype correlation is provided in Table 4.

Real-world clinical behaviour following receipt of BCID2 results

After excluding three patients who died before any antibiotic adjustment could be made and five patients without AST in conventional bacterial cultures, a total of 121 BSI episodes were included in the retrospective analysis on the impact of antimicrobial treatment based on BCID2 results (Fig. 5).

The adequacy of antimicrobial treatment was assessed using conventional culture results with AST. Among the 121 patients, 49 (40.5%) received 'inadequate' empirical antimicrobial treatment. Of the 72 (59.5%) who received an adequate empirical regimen, 41 (33.9%) had a narrower spectrum antibiotic choice.

Following the results of BCID2, 38 patients initially receiving inadequate antimicrobial regimen had their antibiotic adjusted to an adequate one. A total of 25 patients were on adequate empirical antimicrobial regimens, and the BCID2 results confirmed the adequacy of the antibiotic use. Among the 41 patients treated adequately with available narrower-spectrum antibiotics, 4 had their treatment regimen adjusted to another adequate, narrower-spectrum option. In summary, antimicrobial regimen was adjusted or confirmed in 67 (n = 38 + 25 + 4, 55.4%) patients following the results of BCID2.

Table 4 Elaboration on the analysis of the correlation between antimicrobial resistance genotype and phenotype.

Enterobacterales	True positive [ESBL/CTX-M (+)]	False positive [non-ESBL/ CTX-M (+)]	False negative [ESBL/CTX-M (−)]	True negative [non-ESBL/ CTX-M (−)]	Se (%)	Sp (%)	PPV (%)	NPV (%)	Concordance rate (%)
Enterobacter cloacae complex	0	0	0	2	–	100	–	100	100
Escherichia coli	5	0	3	12	63	100	100	80	85
Klebsiella aerogenes	0	0	0	1	–	100	–	100	100
Klebsiella pneumoniae group	0	1	3	10	0	91	0	77	71.4
Proteus spp.	0	0	0	4	–	100	–	100	100
Serratia marcescens	0	0	0	2	–	100	–	100	100
Enterobacterales	True positive [CRE/ Carbapenamase genes (+)]	False positive [non-CRE/ Carbapenamase genes (+)]	False negative [CRE/ Carbapenamase genes (−)]	True negative [non-CRE/ Carbapenamase genes (−)]	Se (%)	Sp (%)	PPV (%)	NPV (%)	Concordance rate (%)
Enterobacter cloacae complex	0	0	1	2	0	100	–	67	66.7
Escherichia coli	1 [a]	0	0	21	100	100	100	100	100
Klebsiella aerogenes	0	0	0	1	–	100	–	100	100
Klebsiella oxytoca	1 [b]	0	0	0	100	–	100	–	100
Klebsiella pneumoniae group	19 [c]	0	0	14	100	100	100	100	100
Proteus spp.	0	0	0	3	–	100	–	100	100
Serratia marcescens	1 [d]	0	0	2	100	100	100	100	100
Gram positive bactereia	True positive [MRSA/*mec*A/C (+)] [VRE/*van*A/B (+)]	False positive [non-MRSA/ *mec*A/C (+)] [non-VRE/ *van*A/B (+)]	False negative [MRSA = *mec*A/C (−)] [VRE = *van*A/B (−)]	True negative [non-MRSA = *mec*A/C (−)] [non-VRE = *van*A/B (−)]	Se (%)	Sp (%)	PPV (%)	NPV (%)	Concordance rate (%)

(*continued on next page*)

Table 4 (*continued*)

Enterobacterales	True positive [ESBL/CTX-M (+)]	False positive [non-ESBL/ CTX-M (+)]	False negative [ESBL/CTX-M (−)]	True negative [non-ESBL/ CTX-M (−)]	Se (%)	Sp (%)	PPV (%)	NPV (%)	Concordance rate (%)
Staphylococcus aureus	5 [e]	0	0	3	100	100	100	100	100
Enterococcus faecium	6 [f]	0	0	0	100	–	100	–	100

[a] KPC ($n = 1$).
[b] VIM ($n = 1$).
[c] KPC ($n = 16$), OXA-48-like ($n = 3$), IMP ($n = 1$), NDM ($n = 1$).
[d] VIM ($n = 1$).
[e] *mecA*/C and MREJ (n = 4), *mecA*/C (n = 2).
[f] *vanA*/B (n = 6).

CRE, Carbapenem-resistant Enterobacterales; ESBL, Extended Spectrum Beta Lactamase; MRSA, Methicillin-resistant *Staphylococcus aureus*; NPV, Negative predictive value; PPV, Positive predictive value; VRE, Vancomycin-resistant Enterococcus.

Discussion

This study explored the clinical application of the BCID2 panel in ICUs experiencing a high burden of drug-resistant pathogens. The BCID2 demonstrates high concordance with conventional cultures results in both bacterial pathogen detection and genotype—phenotype correlation for antimicrobial resistance among common ICU pathogens. Antimicrobial therapy was optimized and confirmed in over 50% of patients after obtaining the BCID2 result. These findings highlight the potential to facilitate rapid treatment and improve outcomes in the ICU with a high burden of drug-resistant pathogens.

In this study, drug-resistant pathogens account for nearly half of the specimens based on conventional culture results, particularly in CRKP (57.1%), *A. calcoaceticus-baumannii* complex (100%), MRSA (70%), and VRE (100%). These findings reveal significantly higher rates compared to those reported in the international EUROBACT-2 study, as well as a national surveillance in this country, the Taiwan Healthcare-associated Infections and Antimicrobial Resistance Surveillance, which reported rates of up to 47.7% carbapenem resistance in *Klebsiella* spp., over 80% carbapenem resistance in *Acinetobacter* spp., as high as 44% for MRSA, and 68% for VRE.[4,15] As drug-resistant pathogens are rapidly rising in the ICU,[4] the recommended practice for antimicrobial therapy before finalizing AST in the ICU is to prescribe empirical antibiotics based on local microbiologic data and risk factors associated with multidrug-resistant (MDR) bacteria.[6] However, no single algorithm can be used to perfectly predict MDR infection.[16] Consequently, the EUROBACT-2 study reported that antimicrobial therapy was considered adequate in only 51.5% within 24 h of blood culture sampling, and antimicrobial resistance lead to a longer delay.[4] Early microbiological diagnosis through molecular rapid diagnostic testing may play a crucial role in initiating timely and appropriate antimicrobial treatment.[8,17]

In this real-world study, the BCID2 Panel achieves rapid diagnosis 40 h faster than conventional culture with complete AST, from the time of blood culture collection to results. Despite variations between studies, when compared to conventional culture methods, BCID2 enables faster identification of BSI pathogens and their resistance genes, facilitating the prompt guidance of semi-targeted antimicrobial therapy.[11,18,19] The BCID2 panel identifies a high proportion of drug resistance genes among common ICU pathogens in this study, especially in 31.9% of Enterobacterales carrying carbapenemase genes. Notably, among all the detected carbapenemase genes, 16.7% were MBLs, indicating potential pan-drug resistance to all available antibiotics in the study hospital. The resistance gene results detected by BCID2, combined with the prevalence of overall resistance, mechanisms of resistance, and the genotype—phenotype correlation of AST, can aid in promptly guiding antimicrobial therapy in the ICU.

The concordance of genotype—phenotype correlation in CRKP, MRSA, and VRE was high, but there was poor correlation between $bla_{\text{CTX-M}}$ and phenotypic ESBL in the *K. pneumoniae* group and *E. coli*. This finding contrasts with a previous study, which found that the BCID2 panel demonstrated high concordance in detecting $bla_{\text{CTX-M}}$ with

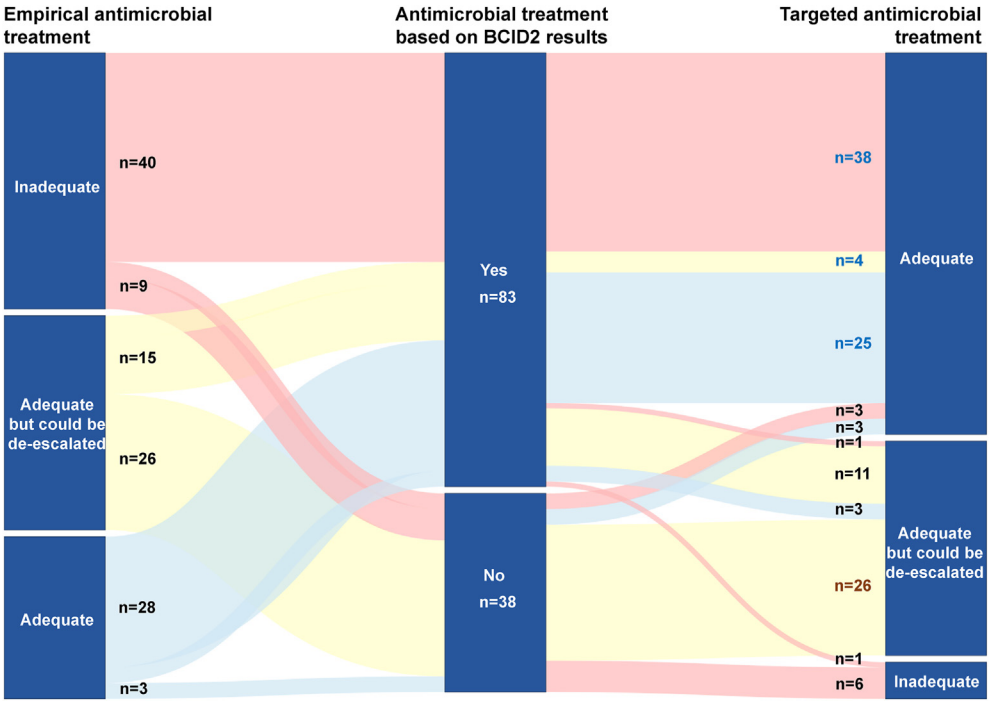

Fig. 5. Sankey diagram of the real-world antimicrobial behavior transitioning from empirical to targeted therapy, guided by the FilmArray® Blood Culture Identification Panel 2 (BCID2) panel. The first column compares the adequacy of empirical antimicrobial treatment to conventional blood culture results with antimicrobial susceptibility testing (AST). The second column represents real-world antimicrobial behavior based on BCID2 results. The third column assesses the adequacy of targeted antimicrobial treatment compared to conventional blood culture results with AST. AST, antimicrobial susceptibility testing.

phenotypic ESBL and considered it the most clinically valuable addition.[10] In this study, the most frequently detected antimicrobial resistant gene in the *K. pneumoniae* group is bla_{KPC}, which is the predominant carbapenemase among carbapenem-non-susceptible *K. pneumoniae* isolates in Taiwan.[20] As discrepancies in prevalence and mechanisms of resistance can result in variations of genotype–phenotype correlation, understanding local epidemiology is important to maintaining the value of mPCR tests.[21] Although mPCR doesn't predict susceptibility or resistance to all antibiotics, the increasing burden of MDRO in the ICU emphasizes the importance of rapidly identifying the pathogen and its resistance genes. In the future, enhancing the accuracy of predicting phenotypic susceptibility and resistance could involve utilizing next-generation sequencing and analyzing full genomic data rather than relying on single gene markers.[22,23]

The BCID2 demonstrated limited sensitivity in identifying *P. aeruginosa*, *E. faecium*, and *S. aureus* in this study, possibly due to constraints in analyzing polymicrobial infections. In our cohort, 26 out of 143 pathogens identified by conventional culture were associated with polymicrobial infections, including seven instances of *P. aeruginosa*, *E. faecium*, and *S. aureus*. Previous research has indicated that the BCID2 panel's efficacy in polymicrobial blood cultures is less convincing.[12,18] An overview of discordant species in polymicrobial blood culture identification by conventional culture and the BCID2 panel is presented in Supplementary Table 2. These findings underscore the importance of cautious interpretation of BCID2 results in polymicrobial infections.

Our data revealed a high proportion (40.5%) of inadequate empirical antimicrobial treatment (IEAT) and BCID2 demonstrated a high impact (55.4%) on antimicrobial stewardship in the medical ICU with a high burden of MDRO. Given that antimicrobial resistance contributes to prolonging the delays in initiating appropriate antimicrobial therapy,[4] the mPCR method enables providers to make more confident antibiotic selections before obtaining full AST results.[22,24,25] In our study, the majority of adjustments involved transitioning from inadequate to adequate antimicrobial treatment based on BCID2 results. It worth nothing that among the patients receiving adequate treatment with available narrower spectrum antibiotics, only four patients de-escalated antimicrobial treatment. Most patients did not de-escalate antimicrobial treatment even when BCID2 detected no pathogens or resistance genes. This finding differs from a randomized trial that showed the use of mPCR tests led to a reduction in unnecessary antibiotic use and facilitated antibiotic de-escalation.[8] The presence of MDRO risk factors and the disease severity was high in this study, which may lead to broad-spectrum antimicrobial treatment and make clinicians hesitant to de-escalate. Additionally, for the optimal impact of mPCR tests, the rapid results should be provided with real-time decision support to aid in the clinical interpretation of mPCR reports and the adjustment of antimicrobial strategies.[8] Fig. S2 provides insights into a strategy for antimicrobial stewardship for BSI based on BCID2, with the aim of rapidly identifying BCID2-detectable pathogens and resistant genes in the ICU and initiating mPCR-based targeted therapy promptly.

This study has several limitations. First, it is a single-center retrospective study with a limited number of cases; a larger sample size from diverse regions is necessary to enhance clinical representativeness. Second, the accuracy of BCID2 performance on rare species in the ICU may be compromised due to a relatively small number of cases. Third, off-panel organisms, which the BCID2 Panel is not designed to detect, were identified by conventional culture in 14.7% (21/143) of our cohort and around 10% in previous studies.[10] However, the numbers of each of these off-panel organisms are too limited to draw any conclusions in this study, and further research is needed to justify their clinical implications. Fourth, the study design excluded blood cultures flagged as potential contamination from BCID2 testing, potentially underestimating the prevalence of common GP pathogens in the ICU, such as *E. faecium* and *S. aureus*. Fifth, the time from sampling to results in our study was longer compared to previous literature.[18,19] In a real-world setting, factors like courier delivery, laboratory processing, and working hours can influence this duration. Nevertheless, our study still highlights BCID2's time-saving benefits for microbiologic diagnosis. Based on the resources of the clinical laboratory, the possibility of running the test 24/7 rather than only during working hours is believed to further impact the timely prescription of antimicrobials. Sixth, due to the retrospective design, researchers did not influence antimicrobial treatment by clinical physicians in this study. Further prospective studies are needed. Seventh, in this study, ESBL was defined phenotypically as resistant to 3rd generation cephalosporins and genotypically as $bla_{CTX\text{-}M}$ detection. However, the presence of the $bla_{CTX\text{-}M}$ gene may not explain all cases of 3rd generation cephalosporin resistance.

Conclusion

In conclusion, this study reveals a high prevalence of drug-resistant pathogens in the ICU, with IEAT administered to 40.5% of patients. The BCID2 panel detect most common pathogens in the ICU more rapidly than conventional culture with complete AST, with high concordance in genotype—phenotype correlation in antimicrobial resistance. BCID2 demonstrated an impact on antimicrobial stewardship in more than 50% of ICU patients. To optimize its utilization and enhance its impact, integrating mPCR results with antimicrobial stewardship programs and real-time decision support is necessary.

Ethics statement

This study received approval from the IRB of the CMUH (CMUH108-REC2-111, CMUH111-REC1-074).

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Declaration of competing interest

The authors declare no conflict of interest.

Data availability statement

The data of this study are available on request from the corresponding author.

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References

1. Kumar A, Roberts D, Wood KE, Light B, Parrillo JE, Sharma S, et al. Duration of hypotension before initiation of effective antimicrobial therapy is the critical determinant of survival in human septic shock. *Crit Care Med* 2006;**34**:1589—96.
2. Sherwin R, Winters ME, Vilke GM, Wardi G. Does early and appropriate antibiotic administration improve mortality in

emergency department patients with severe sepsis or septic shock? *J Emerg Med* 2017;**53**:588—95.
3. De Waele JJ, Akova M, Antonelli M, Canton R, Carlet J, De Backer D, et al. Antimicrobial resistance and antibiotic stewardship programs in the ICU: insistence and persistence in the fight against resistance. A position statement from ESICM/ESCMID/WAAAR round table on multi-drug resistance. *Intensive Care Med* 2018;**44**:189—96.
4. Tabah A, Buetti N, Staiquly Q, Ruckly S, Akova M, Aslan AT, et al. Epidemiology and outcomes of hospital-acquired bloodstream infections in intensive care unit patients: the EUROBACT-2 international cohort study. *Intensive Care Med* 2023;**49**:178—90.
5. Santajit S, Indrawattana N. Mechanisms of antimicrobial resistance in ESKAPE pathogens. *BioMed Res Int* 2016;**2016**:2475067.
6. Sy CL, Chen PY, Cheng CW, Huang LJ, Wang CH, Chang TH, et al. Recommendations and guidelines for the treatment of infections due to multidrug resistant organisms. *J Microbiol Immunol Infect* 2022;**55**:359—86.
7. Collaborators AR. Global burden of bacterial antimicrobial resistance in 2019: a systematic analysis. *Lancet* 2022;**399**:629—55.
8. Banerjee R, Teng CB, Cunningham SA, Ihde SM, Steckelberg JM, Moriarty JP, et al. Randomized trial of rapid multiplex polymerase chain reaction-based blood culture identification and susceptibility testing. *Clin Infect Dis* 2015;**61**:1071—80.
9. Kang CM, Chen XJ, Chih CC, Hsu CC, Chen PH, Lee TF, et al. Rapid identification of bloodstream bacterial and fungal pathogens and their antibiotic resistance determinants from positively flagged blood cultures using the BioFire FilmArray blood culture identification panel. *J Microbiol Immunol Infect* 2020;**53**:882—91.
10. Rhoads DD, Pournaras S, Leber A, Balada-Llasat JM, Harrington A, Sambri V, et al. Multicenter evaluation of the BIOFIRE Blood Culture Identification 2 Panel for detection of bacteria, yeasts, and antimicrobial resistance genes in positive blood culture samples. *J Clin Microbiol* 2023;**61**:e0189122.
11. Peri AM, Ling W, Furuya-Kanamori L, Harris PNA, Paterson DL. Performance of BioFire Blood Culture Identification 2 Panel (BCID2) for the detection of bloodstream pathogens and their associated resistance markers: a systematic review and meta-analysis of diagnostic test accuracy studies. *BMC Infect Dis* 2022;**22**:794.
12. Berinson B, Both A, Berneking L, Christner M, Lutgehetmann M, Aepfelbacher M, et al. Usefulness of BioFire FilmArray BCID2 for blood culture processing in clinical practice. *J Clin Microbiol* 2021;**59**:e0054321.
13. Timsit JF, Ruppé E, Barbier F, Tabah A, Bassetti M. Bloodstream infections in critically ill patients: an expert statement. *Intensive Care Med* 2020;**46**:266—84.
14. Elzi L, Babouee B, Vögeli N, Laffer R, Dangel M, Frei R, et al. How to discriminate contamination from bloodstream infection due to coagulase-negative staphylococci: a prospective study with 654 patients. *Clin Microbiol Infect* 2012;**18**:E355—61.
15. Taiwan C.D.C. Taiwan Healthcare-associated infection and Antimicrobial resistance Surveillance System (THAS). Available from: https://www.cdc.gov.tw/En/Category/Page/J63NmsvevBg2u3I2qYBenw Accessed March, 9, 2024.
16. Timsit JF, Bassetti M, Cremer O, Daikos G, de Waele J, Kallil A, et al. Rationalizing antimicrobial therapy in the ICU: a narrative review. *Intensive Care Med* 2019;**45**:172—89.
17. Giacobbe DR, Giani T, Bassetti M, Marchese A, Viscoli C, Rossolini GM. Rapid microbiological tests for bloodstream infections due to multidrug resistant Gram-negative bacteria: therapeutic implications. *Clin Microbiol Infect* 2020;**26**:713—22.
18. Sparks R, Balgahom R, Janto C, Polkinghorne A, Branley J. Evaluation of the BioFire Blood Culture Identification 2 panel and impact on patient management and antimicrobial stewardship. *Pathology* 2021;**53**:889—95.
19. Peri AM, Bauer MJ, Bergh H, Butkiewicz D, Paterson DL, Harris PN. Performance of the BioFire blood culture identification 2 panel for the diagnosis of bloodstream infections. *Heliyon* 2022;**8**:e09983.
20. Lee YL, Lu MC, Shao PL, Lu PL, Chen YH, Cheng SH, et al. Nationwide surveillance of antimicrobial resistance among clinically important Gram-negative bacteria, with an emphasis on carbapenems and colistin: results from the Surveillance of Multicenter Antimicrobial Resistance in Taiwan (SMART) in 2018. *Int J Antimicrob Agents* 2019;**54**:318—28.
21. Spafford K, MacVane S, Humphries R. Evaluation of empiric β-Lactam susceptibility prediction among Enterobacteriaceae by molecular β-Lactamase gene testing. *J Clin Microbiol* 2019;**57**(10):e00674. -19.
22. Lakbar I, Singer M, Leone M. 2030: will we still need our microbiologist? *Intensive Care Med* 2023;**49**:1232—4.
23. Humphries RM, Bragin E, Parkhill J, Morales G, Schmitz JE, Rhodes PA. Machine-learning model for prediction of Cefepime susceptibility in Escherichia coli from whole-genome sequencing data. *J Clin Microbiol* 2023;**61**:e0143122.
24. Peri AM, Harris PNA, Paterson DL. Culture-independent detection systems for bloodstream infection. *Clin Microbiol Infect* 2022;**28**:195—201.
25. Samuel L. Direct-from-blood detection of pathogens: a review of technology and challenges. *J Clin Microbiol* 2023;**61**:e0023121.

Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.jmii.2024.06.004.