Abstract submitted to ESCMID (Awaiting acceptance) but accepted at ASM Microbe Washington June 2026

From Molecular Diagnostics to Artificial Intelligence: Impact of an Integrated Bloodstream Infection Program on Diagnostic Concordance and Time-to-Therapy

Juan C. Gómez de la Torre [1,2], Ari Frenkel [2], Carlos Chavez-Lencinas [3,4], Alicia Rendon [2], Yoshie Higuchi[1] , Luis Alvarado[1,] Jose Alonso Cáceres-DelAguila[1] and Miguel Hueda-Zavaleta [5,*]

[1]Clinical Laboratory Roe, Lima 15076, Perú; jgomez@labroe.com; jcaceres@labroe.com; Lalvarado@labroe.com; yoshie.higuchi@labroe.com
[2]Arkstone Medical Solutions, Florida, 33428, USA; afrenkel@arkstonemedical.com
[3]Hospital Nacional Edgardo Rebagliati Martins, Lima 15073, carlos.chavez@essalud.gob.pe
[4]Universidad Nacional Mayor de San Marcos, Lima 15072, Perú; cchavezl1@unmsm.edu.pe
[5] Diagnóstico, tratamiento e investigación de enfermedades infecciosas y tropicales, Universidad Privada de Tacna, Tacna 23003, Peru, mighueda@virtual.upt.pe
***Correspondence: mighueda@virtual.upt.pe; Tel.: +51996940584**

ABSTRACT

Purpose: Rapid molecular diagnostics combined with artificial intelligence (AI) clinical decision support systems (CDSS) promise to reduce time to appropriate antimicrobial therapy in bloodstream infections (BSI), but optimal integration with conventional culture remains undefined. We evaluated an integrated diagnostic-therapeutic program combining molecular-AI initial guidance with phenotypic-AI optimization.

Methods: We prospectively analyzed 237 consecutive monomicrobial BSI episodes (September 2023–October 2025) processed through an integrated program utilizing parallel rapid molecular diagnostics (FilmArray BCID or Xpert MRSA/SA BC) with OneChoice® AI recommendations at 19.7 hours, and conventional culture with MALDI-TOF identification plus phenotypic antimicrobial susceptibility testing (AST) with OneChoice Fusion® AI optimization at 52.9 hours. Primary objectives assessed: (1) diagnostic turnaround time reduction; (2) bacterial identification and antimicrobial resistance detection concordance; and (3) therapeutic recommendation concordance.

Results: Molecular diagnostics reduced turnaround time by 33.2±14.2 hours compared to conventional methods (62.7% reduction; $P<0.001$; Cohen's $d=2.33$). Combined concordance rates were: bacterial identification 94.5% (225/237; 95% CI: 90.8-96.8%; Cohen's κ=0.84, almost perfect), antimicrobial resistance 89.5% (212/237; 95% CI: 84.9–92.8%; κ=0.76, substantial), and therapeutic recommendations 81.4% (192/236; 95% CI: 75.9-85.8%; κ=0.55, moderate). Multivariable analysis identified resistance detection discordance as the sole independent predictor of therapeutic discordance (adjusted OR 7.70, 95% CI: 3.61 - 17.49, $P=0.001$). The integrated strategy delivered appropriate therapy 33 hours earlier in 81.4% of cases without requiring adjustment, with phenotypic optimization ensuring 100% appropriate therapy.

Conclusion: An integrated diagnostic-therapeutic program combining molecular-AI initial guidance with phenotypic-AI optimization achieves substantial time reductions while maintaining high diagnostic and therapeutic concordance, supporting implementation in antimicrobial stewardship programs.

INTRODUCTION

Sepsis constitutes a critical global health challenge, with an estimated 48.9 million cases and 11.0 million deaths annually (1). Bloodstream infections (BSI) are a primary cause of sepsis. Despite advancements in supportive care, mortality rates remain substantial, ranging from 30-42%, with half of these fatalities directly attributable to infection (2,3). The clinical imperative is further complicated by escalating antimicrobial resistance (AMR), particularly in resource-constrained settings where resistance rates exceed 50% for common Gram-negative pathogens (4). In Peru, resistance to third-generation cephalosporins reaches 51.7% in *Escherichia coli* and 72.7% in *Klebsiella pneumoniae* bloodstream isolates, significantly surpassing global averages (5).

The axiom "time is life" in sepsis management underscores the critical importance of expeditious pathogen identification and antimicrobial susceptibility determination. Each hour of delay in the administration of appropriate antibiotics is demonstrably associated with increased mortality, with survival rates diminishing by 7.6% per hour in cases of septic shock (6,7). Conventional diagnostic protocols, which rely on blood culture incubation, matrix-assisted laser desorption/ionization time-of-flight mass spectrometry (MALDI-TOF MS) identification, and phenotypic antimicrobial susceptibility testing (AST), typically necessitate 48-72 hours for definitive results (8,9). This protracted timeline mandates empirical therapy, which proves inappropriate in 20-30% of cases, culminating in a 2- to 3-fold increase in mortality and extended hospitalization (10–12). Conversely, the timely initiation of appropriate therapy within the initial hours significantly enhances survival, with a reduction in sepsis mortality observed for each hour gained (13).

Rapid molecular diagnostic platforms, such as multiplex polymerase chain reaction panels (FilmArray® Blood Culture Identification, BioFire Diagnostics) and targeted assays (Xpert® MRSA/SA Blood Culture, Cepheid), have substantially advanced the field of bloodstream infection (BSI) diagnostics. These platforms facilitate pathogen identification and resistance marker detection within 1-2 hours of a positive blood culture signal (14,15). They are capable of identifying resistance genes, including extended-spectrum β-lactamases (ESBL), *mecA* (methicillin resistance), and carbapenemase genes, thereby reducing the diagnostic turnaround time by approximately 24-48 hours compared to conventional methodologies (16). Nevertheless, molecular diagnostics possess inherent limitations: these panels are restricted to the detection of predefined targets, offer genus-level identification without comprehensive susceptibility profiles, and are unable to detect chromosomal resistance mechanisms, such as AmpC β-lactamases in *Enterobacter* spp. (17). Meta-analyses indicate that while rapid molecular diagnostics contribute to a reduction in the time to appropriate therapy, their clinical impact is critically dependent upon

integration with antimicrobial stewardship programs to effectively translate genotypic data into actionable therapeutic decisions (18,19).

Artificial intelligence (AI) and machine learning (ML) technologies have emerged as transformative tools to bridge this translational gap. Clinical Decision Support Systems (CDSS), trained on extensive datasets encompassing molecular and phenotypic microbiology results, are capable of generating evidence-based antimicrobial recommendations in real-time (20,21). Recent validation studies of AI-CDSS platforms demonstrate a high degree of concordance with infectious disease specialist recommendations (>80%) when utilizing molecular data alone, approaching 100% accuracy upon integrating both molecular and phenotypic data (in press). The OneChoice® platform (Arkstone Medical Solutions), a human-in-the-loop AI system validated for antimicrobial stewardship, has exhibited 100% accuracy in novel data prediction and an 84.5% concordance with clinical practice guidelines (22). A preliminary comparative analysis revealed that OneChoice recommendations, when based solely on molecular data, achieved an an 81.4% concordance with those generated using integrated molecular-phenotypic data (OneChoice Fusion), with molecular-guided recommendations becoming available 33.2 hours earlier (23).

Our institution has implemented an integrated bloodstream infection diagnostic and therapeutic program, building upon existing technological advancements. This program combines continuous automated blood culture monitoring, parallel rapid molecular and conventional culture workflows, and dual AI-guided antimicrobial recommendations at both molecular and phenotypic data integration points. The program operates as follows: upon a positive blood culture signal (typically occurring within 12-24 hours), Gram staining directs parallel processing. This involves (1) rapid molecular testing (FilmArray BCID or Xpert MRSA/SA BC), which generates pathogen identification and select resistance markers within 1-2 hours, coupled with OneChoice AI analysis providing initial therapeutic recommendations approximately 19-20 hours from blood draw. Concurrently, (2) conventional culture on solid media is performed for MALDI-TOF identification and comprehensive phenotypic antimicrobial susceptibility testing (AST), generating definitive susceptibility data approximately 48-60 hours, which is then integrated by OneChoice Fusion to provide optimized therapy recommendations. This integrated approach theoretically enables early targeted therapy while maintaining phenotypic confirmation for therapeutic optimization; however, the extent to which molecular-AI recommendations reliably guide initial therapy across diverse pathogens and resistance phenotypes necessitates rigorous evaluation.

This study aimed to thoroughly assess the clinical utility of an integrated diagnostic-therapeutic program within a real-world cohort of patients afflicted with bacteremia and fungemia. Our specific objectives were to ascertain: (1) the reduction in diagnostic turnaround time achieved by molecular methodologies in contrast to conventional culture-based workflows; (2) the diagnostic concordance for bacterial identification and antimicrobial resistance detection between molecular and conventional methods; and (3) the therapeutic concordance between molecular-AI–guided recommendations (OneChoice) and integrated molecular-phenotypic-AI recommendations (OneChoice Fusion), while also identifying predictors of discordance and characterizing pathogen-specific performance profiles.

MATERIALS AND METHODS

Study Design and Setting

This prospective observational study was conducted from September 2023 through October 2025 at a tertiary-care reference laboratory in Peru, serving multiple medical specialties including internal medicine, emergency medicine, intensive care, nephrology, and hematology. The study protocol received approval from the institutional ethics committee (IRB approval) and adhered to the Declaration of Helsinki. Written informed consent was waived for this observational study, which utilized de-identified clinical microbiology data generated during routine patient care.

Study Population

All consecutive patients presenting with positive blood cultures, processed via the integrated bloodstream infection (BSI) diagnostic program during the study period, were deemed eligible for inclusion. Eligibility criteria encompassed: (1) mono or polymicrobial bacteremia or fungemia substantiated by positive blood culture; (2) comprehensive diagnostic testing conducted through both rapid molecular and conventional culture methodologies; (3) the availability of AI-generated therapeutic recommendations from both the OneChoice® (molecular-based) and OneChoice Fusion® (integrated molecular-phenotypic) platforms; and (4) complete clinical and microbiological data. Exclusion criteria consisted of cases characterized by incomplete diagnostic or therapeutic data. The final cohort comprised 237 BSI episodes from distinct patients.

Integrated BSI Diagnostic and Therapeutic Program

Our institution has implemented a standardized integrated diagnostic-therapeutic program combining continuous automated blood culture monitoring, parallel rapid molecular and conventional culture workflows, and dual AI-guided antimicrobial decision support, as previously described (30,31). The program workflow is summarized in Figure 1 and detailed below.

Blood Culture Processing

Blood cultures were collected following institutional protocols for aseptic sampling and inoculated into automated continuous monitoring blood culture bottles. Samples were incubated in automated blood culture systems with continuous colorimetric or fluorometric detection of bacterial growth. Upon positive signal detection (typically 12-24 hours from blood draw; range 8-36 hours), bottles were immediately removed for parallel processing through molecular and conventional pathways.

Rapid Molecular Diagnostic Pathway

Gram Staining: Positive blood culture bottles underwent immediate Gram staining (within 1 hour of positive signal) to determine bacterial morphology: Gram-negative bacilli, Gram-positive bacilli, Gram-positive cocci in chains, Gram-positive cocci in clusters, or yeast. Gram stain morphology directed subsequent molecular test selection.

Molecular Panel Testing: Samples were processed using one of two FDA-cleared molecular platforms based on Gram stain morphology:

- FilmArray® Blood Culture Identification (BCID) Panel (BioFire Diagnostics, Salt Lake City, UT, USA) for Gram-negative bacilli, Gram-positive bacilli, yeasts, and Gram-positive cocci in chains. This nested multiplex PCR panel detects 24 bacterial and 5 yeast species, plus 3 resistance genes (*CTX-M, IMP, KPC, NDM, OXA-48-like, VIM, mcr-1, mecA/C, mecA/C + MREJ,* y *vanA/B*.), with results available within 1 hour.
- Xpert® MRSA/SA Blood Culture (Cepheid, Sunnyvale, CA, USA) for Gram-positive cocci in clusters, specifically targeting *Staphylococcus aureus* identification and *mecA* gene detection, with results within 1 hour. This molecular testing was prioritized for positive signals occurring within 16 hours of incubation, as this time threshold has been validated as predictive of pathogenic organisms and optimizes resource utilization.

OneChoice® AI Analysis (Molecular-Based Recommendations): Molecular identification and resistance gene results were automatically transmitted to the OneChoice® platform (Arkstone Medical Solutions, USA), a machine learning-based clinical decision support system trained on FDA antimicrobial indications, IDSA clinical practice guidelines, meta-analyses, and local epidemiological data. The system integrates patient-specific factors (age, pregnancy status, documented allergies, renal function) and institutional formulary constraints to generate evidence-based antimicrobial recommendations. Recommendations were transmitted to treating physicians via secure electronic reporting within 1 hour of molecular result availability. The validation of this MLHL software has already been published (22). Supplement 1

Conventional Culture and Phenotypic Pathway

Conventional Culture: In parallel with molecular testing, positive blood culture samples were subcultured onto solid media (blood agar, MacConkey agar, and chocolate agar) and incubated at 35°C in ambient air or 5% CO_2, as appropriate for the organism's growth. Pure colonies were harvested after 24-48 hours for identification and susceptibility testing.

MALDI-TOF MS Identification: Bacterial and fungal identification was performed using VITEK® MS (bioMérieux, Marcy-l'Étoile, France) following manufacturer protocols. Briefly, direct colony smears were prepared, overlaid with α-cyano-4-hydroxycinnamic acid matrix, and analyzed by matrix-assisted laser desorption/ionization time-of-flight mass spectrometry. Identifications with high confidence level (confidence value 60 - 99.9) were accepted as species-level identification; Identifications with medium medium confidence level underwent repeat testing or alternative identification methods.

Phenotypic Antimicrobial Susceptibility Testing: Comprehensive AST was performed using the VITEK® 2 Compact automated system (bioMérieux, Marcy-l'Étoile, France) with organism-specific antimicrobial panels (AST-N403 for Gram-negatives, AST-P663 or AST-ST03 for Gram-positives, AST-YS08 for yeasts). Minimum inhibitory concentrations (MIC) were interpreted using Clinical and Laboratory Standards Institute (CLSI) M100 breakpoints (35th edition, 2025)(35). Suspected ESBL-producing *Enterobacterales* underwent confirmatory testing; cefoxitin resistance patterns identified suspected inducible AmpC producers.

OneChoice Fusion® AI Analysis (Integrated Recommendations): Upon completion of MALDI-TOF identification and phenotypic AST, comprehensive microbiological data

(organism identification, complete antibiogram, genotypic resistance markers from molecular testing, phenotypic resistance mechanisms) were transmitted to the OneChoice Fusion® platform. This integrated AI system re-analyzes all available data (molecular genotypic, conventional phenotypic, and patient-specific clinical factors) to generate optimized therapeutic recommendations, including dosing adjustments based on pharmacokinetic/pharmacodynamic principles and patient renal function. Fusion recommendations were transmitted to physicians within 2 hours of completing the phenotypic AST. Supplement 2

Data Collection and Variables

For each BSI episode, we collected: (1) patient demographics (age, sex); (2) clinical setting (hospitalized vs. community-acquired, source department); (3) time metrics (blood draw to positive signal, positive signal to molecular result, positive signal to phenotypic AST result, total turnaround times for each pathway); (4) microbiological results (organism identified by molecular panel, organism identified by MALDI-TOF, resistance genes detected, phenotypic resistance patterns, antibiogram results); and (5) AI-generated therapeutic recommendations (OneChoice molecular-based recommendation, OneChoice Fusion integrated recommendation).

Concordance Definitions

Bacterial Identification Concordance: Concordance between molecular and MALDI-TOF identification was classified as: (1) complete concordance = identical species-level identification; (2) partial concordance = molecular panel identified genus only while MALDI-TOF provided genus and species; (3) discordance = different organisms identified, including cases where the molecular panel failed to detect pathogens outside its target range or MALDI-TOF identified contaminants.

Antimicrobial Resistance Concordance: Concordance between genotypic (molecular gene detection) and phenotypic (AST) resistance was classified as: (1) complete concordance = resistance gene detection correlated with phenotypic resistance to corresponding antimicrobial class; (2) partial concordance = minor discrepancies not affecting therapeutic decisions; (3) discordance = gene-positive/phenotype-susceptible or gene-negative/phenotype-resistant discrepancies, primarily occurring with chromosomal mechanisms (e.g., AmpC) not detected by molecular panels.

Therapeutic Concordance: Concordance between OneChoice and OneChoice Fusion recommendations was classified as: (1) concordant = identical antimicrobial agent, dosage, and route recommended by both platforms; (2) discordant = different antimicrobial recommendations, requiring therapeutic adjustment based on phenotypic data.

Statistical Analysis

Continuous variables were assessed for normality using the Shapiro-Wilk test and expressed as mean ± standard deviation (SD) or median with interquartile range (IQR) as appropriate. Categorical variables were expressed as frequencies and percentages. Diagnostic turnaround times for molecular versus conventional methods were compared using the Wilcoxon signed-rank test for paired data. Effect size was quantified using Cohen's *d* for paired samples. Concordance proportions were calculated with 95% confidence intervals (CI) using the Wilson score method. Agreement beyond chance was assessed using Cohen's kappa (κ) coefficient, with

interpretation according to Landis and Koch criteria (κ<0.20 poor, 0.21-0.40 fair, 0.41-0.60 moderate, 0.61-0.80 substantial, >0.80 almost perfect).

Univariable and multivariable logistic regression analyses were performed to identify predictors of therapeutic discordance, with results expressed as odds ratios (OR) with 95% CI. Variables with *P*<0.10 in univariable analysis were entered into multivariable models. Subgroup analyses were performed by stratifying according to Gram stain classification, resistance presence, clinical setting, and specific pathogens. Between-group comparisons utilized the Chi-square test or Fisher's exact test for categorical variables and the Mann-Whitney U test for continuous variables. Statistical significance was defined as two-tailed *P*<0.05. For Figures 4 and 5, PDF AI and Cloud 4.5 were used to generate the images based on the database, with information corroborated by manual methods and STATA 16 software. The remaining graphs and tables were created using RStudio 2025.05.0 software.

RESULTS

Study Population and Baseline Characteristics

During the study period (September 2023 to October 2025), 237 bloodstream infection episodes from unique patients met inclusion criteria and comprised the final analytical cohort. Patient demographics and baseline characteristics are summarized in **Table 1.** The cohort's mean age was 58.3 ± 26.5 years (median, 64 years; range, 0-100 years), with males comprising 56.1% (133/237) of the participants. Clinical settings included outpatient/community settings (51.1%, 121/237), hospitalized patients (47.7%, 113/237), and emergency department presentations (10.9%, 26/237).

The pathogen distribution reflected typical BSI epidemiology with predominance of Gram-negative bacteria **(Figure 2)**. *Escherichia coli* was the most frequently isolated organism (35.0%, 83/237), followed by *Klebsiella pneumoniae* (10.5%, 25/237), *Salmonella* spp. (7.2%, 17/237), *Streptococcus* spp. (6.8%, 16/237), *Staphylococcus aureus* (6.3%, 15/237), and *Enterobacter* spp. (6.3%, 15/237). Gram-negative bacteria accounted for 73.4% (174/237) of the isolates, Gram-positive bacteria for 19.0% (45/237), fungi for 3.4% (8/237), and molecular panels failed to detect pathogens in 3.4% (8/237) of cases (organisms outside the panel targets). Antimicrobial resistance was detected in 43.0% (102/237) of isolates, including 21.9% (52/237) with ESBL production (predominantly CTX-M), 10.1% (24/237) with chromosomal AmpC,0.42% (1/237) with MRSA (*mecA*-positive), and 1.26% (3/237) with carbapenemases.

Primary Objective 1: Reduction in Diagnostic Turnaround Time

Complete timing data for both molecular and conventional diagnostic pathways were available for 237 BSI episodes (100% of the cohort). The distribution of turnaround times and time savings is illustrated in **Figures 3 and 4**. Molecular diagnostics yielded results in a mean of 19.7 ± 10.9 hours (median 16.4 hours; IQR 14.1-20.6 hours; range 6.7-67.0 hours) from blood draw. Conventional culture with MALDI-TOF identification and phenotypic AST required a mean of 52.9 ± 19.4 hours (median 47.5 hours; IQR 41.4-61.4 hours; range 23.2-134.0 hours). The mean time reduction achieved by molecular diagnostics was 33.2 ± 14.2 hours (median 30.7 hours; IQR 24.6-38.9 hours), representing a 62.7% reduction in diagnostic turnaround time (Wilcoxon signed-rank test: *P*<0.001; Cohen's *d*=2.33, considerable effect size).

Subgroup analysis revealed consistent and highly significant time savings across all clinical categories (**Table 2**). Time reduction was similar between Gram-negative (30.6

± 11.1 hours, *P < 0.001) and Gram-positive (39.3 ± 10.5 hours, P < 0.001) bacterial infections, with no significant difference between the groups (Mann-Whitney U test: P = 0.081*). Fungi demonstrated the longest turnaround times for both molecular (37.3 ± 16 hours) and conventional methods (92.7 ± 43.1 hours), with substantial time savings (55.3 ± 42.4 hours); with statistical significance (*n* = 8, *P* = 0.008). Time savings did not differ significantly between cases with antimicrobial resistance (30.8 ± 11.6 hours) and susceptible isolates (38.8 ± 15.7 hours, *P* = 0.238), nor between hospitalized (33.4 ± 12.8 hours) and outpatient settings (32.7 ± 15.4 hours, *P* = 0.921). Pathogen-specific analysis demonstrated consistent time reductions across major pathogens: *E. coli* (28.0 ± 8.1 hours), *K. pneumoniae* (31.3 ± 14.9 hours), *S. aureus* (33.6 ± 6.9 hours), *P. aeruginosa* (32.9 ± 12.8 hours), and *Salmonella* spp. (27.4 ± 8.9 hours), with all comparisons achieving *P*<0.001.

Primary Objective 2: Diagnostic Concordance

Bacterial Identification Concordance

Bacterial identification concordance between molecular panels and MALDI-TOF MS was evaluable in all 237 cases (**Table 2A**). Complete species-level concordance was achieved in 189 cases (79.7%), partial concordance (molecular genus-level identification, MALDI-TOF species-level identification) in 35 cases (14.8%), and discordance in 13 cases (5.1%). Combined concordance (complete plus partial) was 94.5% (225/237; 95% CI: 91.4-97.2%; Cohen's κ=0.84, almost perfect agreement).

Subgroup analysis revealed heterogeneity in concordance patterns by pathogen and clinical characteristics (**Table 2A**). Gram-negative bacteria demonstrated excellent combined concordance (95.5%, 171/179; 95% CI: 91.4-99.7%), with the majority (83.2%, 149/179) achieving complete species-level agreement. Gram-positive bacteria showed similarly high combined concordance (91.8%, 43/49; 95% CI: 80.8-96.8%) but with greater reliance on partial concordance (26.5%, 13/49), reflecting molecular panels' limitation to genus-level identification for streptococci and enterococci. All fungal isolates (*Candida* spp.) demonstrated complete concordance (100%, 8/8). Pathogen-specific analysis revealed near-perfect concordance for *E. coli* (98.8%, 82/83; κ = 0.84), *K. pneumoniae* (87.0%, 20/23; κ = 0.84), and *S. aureus* (100%, 13/13; κ = 1). *Salmonella* spp. demonstrated 100% combined concordance (17/17) but predominantly as partial concordance (88.2%, 15/17) due to molecular panels identifying only to genus level. The 12 discordant cases comprised organisms outside molecular panel targets (5 cases: *Aeromonas* spp., *Stenotrophomonas maltophilia*, *Raoultella* spp.) or polymicrobial infections detected by culture but reported as monomicrobial by molecular panels (7 cases).

Antimicrobial Resistance Concordance

Antimicrobial resistance concordance between genotypic (molecular gene detection) and phenotypic (AST) testing was evaluable in all 237 cases (**Table 2B**). Complete genotype-phenotype concordance was observed in 210 cases (88.6%), partial concordance in 2 cases (0.8%), and discordance in 25 cases (10.5%). Combined concordance was 89.5% (212/237; 95% CI: 84.9-92.8%; Cohen's κ=0.76, substantial agreement).

Resistance concordance varied by bacterial group and mechanism. Gram-negative bacteria showed high concordance (91.6%, 164/179; κ=0.84), while Gram-positive bacteria (83.7%, 41/49; κ=0.75) and fungi (87.5%, 7/8) demonstrated slightly lower rates. Concordance did not differ significantly between cases with detected resistance

(88.2%, 90/102) versus susceptible isolates (90.4%, 122/135, Chi-square *P*=0.448). Pathogen-specific analysis revealed excellent concordance for *E. coli* (96.4%, 80/83; κ=0.84), *K. pneumoniae* (95.6%, 22/23; κ=0.84), and *S. aureus* (100%, 13/13; κ=0.84).

The 25 discordant cases were categorized into two groups: gene-positive/phenotype-susceptible (16 cases, 64.0%) and gene-negative/phenotype-resistant (9 cases, 36.0%). Gene-positive/phenotype-susceptible discordances involved CTX-M detection with phenotypic cephalosporin susceptibility (2 cases), likely representing low-level ESBL expression or borderline MIC values near breakpoints. Gene-negative/phenotype-resistant discordances primarily involved chromosomal AmpC β-lactamases in *Enterobacter* spp. (7 cases) and *Serratia marcescens* (2 cases), resistance mechanisms not targeted by molecular panels.

Primary Objective 3: Therapeutic Concordance

Overall Therapeutic Concordance

Therapeutic recommendations were available from both OneChoice® (molecular-based) and OneChoice Fusion® (integrated molecular-phenotypic) platforms for 236 cases (99.6% of cohort). Concordant recommendations (identical antimicrobial agent, dosage, and route) were observed in 192 cases (81.4%; 95% CI: 75.9-85.8%; Cohen's κ=0.55, moderate agreement), while discordant recommendations requiring therapeutic adjustment based on phenotypic data occurred in 44 cases (18.6%) (**Table 4**).

Subgroup analysis demonstrated significant variation in therapeutic concordance by clinical characteristics (**Table 4**). Gram-negative infections showed higher concordance (87.4%, 151/179) compared to Gram-positive infections (70.8%, 34/48, Chi-square *P*=0.008). Therapeutic concordance was influenced by upstream diagnostic concordance: cases with bacterial identification concordance demonstrated 86.2% (163/189) therapeutic concordance, while bacterial ID discordant cases showed only 61.7% (29/47) therapeutic concordance (Chi-square *P*<0.001). Similarly, resistance detection concordance strongly influenced therapeutic concordance: resistance-concordant cases achieved 88.0% (184/209) therapeutic concordance versus 29.6% (8/27) in resistance-discordant cases (Chi-square *P*<0.001). Pathogen-specific concordance was highest for *E. coli* (95.1%, 79/83), *S. aureus* (100%, 13/13), and *K. pneumoniae* (91.3%, 23/25).

Analysis of Therapeutic Discordances

Among the 44 therapeutically discordant cases, the underlying reasons for recommendation differences were distributed as follows: resistance detection discordance (34.0%, 15/44), bacterial identification discordance (59%, 26/44), both identification and resistance discordant (18.2%, 8/44), and phenotypic susceptibility-based optimization despite diagnostic concordance (22.7%, 10/44). The nature of therapeutic adjustments included: escalation to broader-spectrum agents based on phenotypic resistance not predicted by genotype (34.1%, 15/44), de-escalation to narrow-spectrum agents based on confirmed susceptibility (27.3%, 12/44), alternative agent selection targeting pathogens outside molecular panel (20.5%, 9/44), and modification based on comprehensive antibiogram availability (18.2%, 8/44).

Predictors of Therapeutic Discordance

Multivariable logistic regression analysis identified resistance detection discordance as the sole independent predictor of therapeutic discordance (adjusted OR 7.70, 95% CI: 3.61-17.49, *P*=0.001) (**Table 5**). Cases with genotype-phenotype resistance discordance had nearly 5-fold increased odds of requiring therapeutic adjustment compared to resistance-concordant cases. Gram-positive bacteria demonstrated a trend toward higher discordance (adjusted OR 1.77, 95% CI: 0.84-3.56, *P*=0.124) but did not reach statistical significance. Other variables, including age, sex, hospitalization status, overall resistance presence, bacterial identification concordance, and molecular turnaround time, were not significant predictors in multivariable analysis.

Integrated Program Performance Summary

The integrated diagnostic-therapeutic program, combining rapid molecular diagnostics with OneChoice AI initial guidance and conventional diagnostics with OneChoice Fusion optimization, achieved the following performance profile (**Figure 5**): (1) 81.4% of patients received appropriate antimicrobial therapy 33.2 hours earlier than conventional workflows would allow, without requiring subsequent adjustment; (2) 18.6% of patients received initial empirical coverage based on molecular-AI guidance, followed by phenotype-guided optimization at 52.9 hours, still benefiting from early pathogen-directed therapy; and (3) 100% of patients ultimately received appropriate personalized antimicrobial therapy informed by comprehensive microbiological data. Among the 192 concordant cases requiring no adjustment, the median time to appropriate therapy was 19.7 hours. Among the 44 discordant cases requiring optimization, initial molecular-AI guidance provided pathogen-directed coverage (though suboptimal agent) at 19.7 hours, with definitive optimal therapy implemented at 52.9 hours, representing substantial improvement over purely empirical therapy pending culture results at 52.9 hours.

DISCUSSION

This prospective observational study evaluated the clinical utility of an integrated bloodstream infection diagnostic-therapeutic program combining rapid molecular diagnostics with dual AI-guided antimicrobial recommendations. Our findings demonstrate that this integrated approach achieves substantial reductions in diagnostic turnaround time (33.2 hours, 62.7% reduction) while maintaining high concordance rates for bacterial identification (94.5%), antimicrobial resistance detection (89.5%), and therapeutic recommendations (81.4%). These results indicate that 81.4% of patients could receive appropriate personalized antimicrobial therapy more than one day earlier than with conventional approaches alone, while the remaining 18.6% receive initial pathogen-directed coverage followed by phenotypic optimization, ensuring 100% eventual appropriate therapy.

The 33.2-hour reduction in diagnostic turnaround time represents one of the most substantial time advantages reported for rapid diagnostic testing in bloodstream infections, with highly significant statistical support (Wilcoxon P<0.001, Cohen's d=2.33). This finding aligns with prior meta-analyses demonstrating that molecular rapid diagnostics reduce time to pathogen identification and resistance characterization(15). However, our study extends beyond diagnostic performance by integrating AI-driven clinical decision support to translate microbiological data into

actionable therapeutic recommendations, addressing the well-documented "interpretation gap" between test results and clinical implementation (24).

The clinical significance of this time advantage cannot be overstated. Multiple landmark studies have demonstrated that each hour of delay in appropriate antimicrobial therapy increases mortality in sepsis and bacteremia by approximately 4-7.6%, with greatest impact within the first 24-48 hours (6,13,25). While our study did not directly measure clinical outcomes, applying evidence from Timbrook et al.'s meta-analysis of 31 studies (n=5,920 patients) to our observed time advantage yields projected mortality reductions from 18.0% to 12.7% (absolute reduction 5.3 percentage points, OR 0.66), corresponding to an estimated 53 lives saved per 1,000 bloodstream infections (number needed to treat=19) (18). Conservative time-based modeling assuming 0.1% mortality reduction per hour suggests at least 27 deaths prevented per 1,000 cases. While these projections require prospective validation, they provide a quantitative framework for understanding potential clinical impact. Beyond mortality, published data suggest the observed time advantage could translate to approximately 1.2 days reduced hospital length of stay per patient, with substantial cost implications (18,26).

Our study demonstrated high combined concordance rates for both bacterial identification (94.5%, κ=0.84, almost perfect agreement) and antimicrobial resistance detection (89.5%, κ=0.76, substantial agreement) between molecular and conventional methods. These results align with prior evaluations of molecular panels, which have reported identification concordance rates of 85-98% depending on panel composition and reference methods (16). The 5.1% bacterial identification discordance was primarily attributable to pathogens outside current molecular panel targets (Aeromonas spp., Raoultella spp.) and polymicrobial infections, collectively representing 5% of cases.

The 10.5% resistance discordance comprised two patterns: gene-positive/phenotype-susceptible (64% of discordances), predominantly CTX-M detection without phenotypic ESBL expression, a documented phenomenon related to heteroresistance, low bacterial inoculum, specific growth conditions, or gene silencing (27,28); and gene-negative/phenotype-resistant (36% of discordances), primarily chromosomal AmpC β-lactamases in Enterobacter spp. not targeted by molecular panels. These discordance patterns highlight fundamental differences between genotypic and phenotypic resistance testing: molecular methods detect the genetic potential for resistance, while phenotypic susceptibility testing measures the actual expression of resistance under standardized laboratory conditions (29,30). Neither approach alone provides complete information, supporting our integrated strategy that leverages molecular diagnostics for early risk stratification, complemented by phenotypic testing for definitive optimization.

The 81.4% therapeutic concordance rate between molecular-AI (OneChoice) and integrated molecular-phenotypic-AI (OneChoice Fusion) recommendations represents a key finding with direct clinical implications. This high concordance indicates that in more than four out of five cases, initial therapy based on rapid molecular diagnostics alone was appropriate without requiring subsequent adjustment when complete phenotypic data became available 33 hours later. Concordance was higher for Gram-negative infections (87.4%) than Gram-positive infections (70.8%, P=0.008), likely reflecting more comprehensive resistance gene coverage for Gram-negatives in current molecular panels. Pathogen-specific concordance was highest for E. coli (95.1%), S. aureus (100%), and K. pneumoniae (91.3%), which collectively represented 52% of our cohort and account for substantial proportions of bloodstream

infections globally (1,2). These high concordance rates for common pathogens suggest molecular-AI guided therapy can confidently replace empirical broad-spectrum therapy in the majority of cases.

Multivariable logistic regression identified resistance detection discordance as the sole independent predictor of therapeutic discordance (adjusted OR 7.70, 95% CI: 3.61-17.49, *P*=0.001). This finding underscores that accurate resistance characterization—rather than bacterial identification per se—is the critical determinant of appropriate antimicrobial selection, emphasizing the importance of comprehensive resistance gene coverage in molecular panels (20,21). The specific reasons for therapeutic discordance provide insights for optimization: 38.6% involved resistance detection discordance, 20.5% bacterial identification discordance, 18.2% both factors simultaneously, and 22.7% represented phenotypic susceptibility-based optimization despite accurate pathogen and resistance identification. These latter cases represent opportunities for therapeutic refinement (e.g., narrow-spectrum alternatives with superior activity) rather than correction of inappropriate initial therapy, underscoring the value of phenotypic testing for antimicrobial optimization even when molecular diagnostics provide accurate initial information.

Our findings support a sequential diagnostic-therapeutic strategy that optimizes both speed and accuracy: rapid molecular diagnostics with AI-guided initial therapy within 19.7 hours, followed by conventional culture with phenotypic susceptibility testing and AI-guided optimization at 52.9 hours. This integrated approach provides 81.4% of patients with appropriate personalized therapy 33.2 hours earlier than conventional methods alone, while ensuring the remaining 18.6% receive pathogen-directed initial coverage followed by definitive optimization. This strategy contrasts favorably with three alternatives: conventional diagnostics alone (delays all patients to 52.9 hours), molecular diagnostics alone (rapid but accepts 18.6% suboptimal therapy), and empirical therapy pending culture results (risks inappropriate coverage, particularly problematic in high-resistance settings) (19,31).

The clinical value of this integrated strategy is particularly evident in high-resistance environments such as our study site in Peru, where third-generation cephalosporin resistance reaches 51.7-72.7% in common Gram-negative bloodstream pathogens (6). In such settings, empirical therapy often requires broad-spectrum agents including carbapenems, contributing to escalating resistance through collateral selection pressure (32,33). Molecular-AI diagnostics enable earlier targeted therapy, reducing unnecessary broad-spectrum exposure while maintaining high rates of appropriate coverage.

Several limitations warrant consideration. First, this single-center study in Peru may not generalize to other settings with different pathogen distributions or resistance patterns, necessitating multicenter validation. Second, we did not directly measure clinical outcomes (mortality, length of stay, complications); while literature-based projections suggest substantial impact, prospective clinical trials are needed for validation. Third, we evaluated specific platforms (FilmArray, Xpert, OneChoice/Fusion); findings may not fully apply to other molecular panels or AI systems. Fourth, we excluded polymicrobial bacteremia (~5-8% of cases), an important scenario warranting dedicated investigation. Fifth, concordance analyses did not include independent clinical adjudication by infectious diseases specialists. Finally, we did not assess actual clinician adherence to AI recommendations or treatment administered.

Future research should include prospective randomized controlled trials measuring clinical outcomes (mortality, morbidity, costs), multicenter validation across diverse settings, expansion of molecular panel coverage for additional resistance mechanisms and rare pathogens, evaluation of metagenomic sequencing as comprehensive alternatives, implementation science studies optimizing clinical workflow integration, and health economic analyses quantifying cost-effectiveness.

Supplementary Materials: The following supporting information can be downloaded at:, Supplement 1: Onechoice Report; and Supplement 2: Onechoice Fusion report.

Author Contributions: Conceptualization, J.C.G.d.l.T. and F.A.; methodology, J.C.G.d.l.T., YH; M.H.-Z.; software, J.C.G.d.l.T., YH .; validation, J.C.G.d.l.T., A.R., C.Ch.L. and YH.; formal analysis, J.C.G.d.l.T., and M.H.-Z.; investigation, J.C.G.d.l.T., F.A., YH and A.R.; resources, J.C.G.d.l.T., YH, C.Ch.L. and M.H.Z.; data curation, J.C.G.d.l.T., C.Ch.L.; writing—original draft preparation, J.C.G.d.l.T. and C.Ch.L. writing—review and editing, J.C.G.d.l.T., F.A., A.R., and M.H.-Z.; visualization, J.C.G.d.l.T., and M.H.-Z.; supervision, F.A. and A.R.; project administration, J.C.G.d.l.T. and F.A.; funding acquisition, F.A. All authors have read and agreed to the published version of the manuscript.

Funding: This research received no external funding.

Institutional Review Board Statement: The study was conducted in accordance with the Declaration of Helsinki and was approved by the Institutional Review Board of the Universidad Privada de Tacna (FACSA-CEI_28-10-2025).

Informed Consent Statement: All the participants accept a CI in each survey

Data Availability Statement: The data analyzed in this manuscript, as well as its definitions, can be downloaded at the following link: Data Base Bacteriemia

Acknowledgments: We thank all personnel at Arkstone Medical Solutions and Roe Clinical Laboratory who have been actively working.

Conflicts of Interest: Ari Frenkel is Chief Science Officer of Arkstone Medical Solutions, the company that produces the OneChoice report evaluated in this study. JC Gómez de la Torre works as the Director of Molecular Informatics at Arkstone Medical Solutions and as the Medical Director at Roe Lab in Perú. Yoshie Huguchi works at Roe Laboratory. At the same time, Alicia Rendon, Carlos Chavez L., and Miguel Hueda Zavaleta serve as Quality Assurance Managers at Arkstone Medical Solutions. These affiliations may be perceived as potential conflicts of interest. However, the study's design, data collection, analysis, interpretation, manuscript preparation, and the decision to publish the results were conducted independently, with no undue influence from the authors' affiliations or roles within the company.

Bibliography

1. Rudd KE, Johnson SC, Agesa KM, Shackelford KA, Tsoi D, Kievlan DR, et al. Global, regional, and national sepsis incidence and mortality, 1990–2017: analysis for the Global Burden of Disease Study. The Lancet. 2020 Jan;395(10219):200–11.

2. Timsit JF, Ruppé E, Barbier F, Tabah A, Bassetti M. Bloodstream infections in critically ill patients: an expert statement. Intensive Care Med. 2020 Feb;46(2):266–84.

3. Bryan CS. Clinical implications of positive blood cultures. Clin Microbiol Rev. 1989 Oct;2(4):329–53.

4. Palacios-Baena ZR, Giannella M, Manissero D, Rodríguez-Baño J, Viale P, Lopes S, et al. Risk factors for carbapenem-resistant Gram-negative bacterial infections: a systematic review. Clin Microbiol Infect. 2021 Feb 1;27(2):228–35.

5. Rondon C, Garcia C, Krapp F, Machaca I, Olivera M, Fernández V, et al. Antibiotic point prevalence survey and antimicrobial resistance in hospitalized patients across Peruvian reference hospitals. J Infect Public Health. 2023 Dec 1;16:52–60.

6. 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 June;34(6):1589.

7. Evans L, Rhodes A, Alhazzani W, Antonelli M, Coopersmith CM, French C, et al. Surviving sepsis campaign: international guidelines for management of sepsis and septic shock 2021. Intensive Care Med. 2021 Nov 1;47(11):1181–247.

8. Opota O, Croxatto A, Prod'hom G, Greub G. Blood culture-based diagnosis of bacteraemia: state of the art. Clin Microbiol Infect. 2015 Apr 1;21(4):313–22.

9. Patel R. MALDI-TOF MS for the Diagnosis of Infectious Diseases. Clin Chem. 2015 Jan 1;61(1):100–11.

10. Kang CI, Kim SH, Park WB, Lee KD, Kim HB, Kim EC, et al. Bloodstream Infections Caused by Antibiotic-Resistant Gram-Negative Bacilli: Risk Factors for Mortality and Impact of Inappropriate Initial Antimicrobial Therapy on Outcome. Antimicrob Agents Chemother. 2005 Feb;49(2):760–6.

11. Kollef MH, Sherman G, Ward S, Fraser VJ. Inadequate Antimicrobial Treatment of Infections: A Risk Factor for Hospital Mortality Among Critically Ill Patients. CHEST. 1999 Feb 1;115(2):462–74.

12. Paul M, Shani V, Muchtar E, Kariv G, Robenshtok E, Leibovici L. Systematic Review and Meta-Analysis of the Efficacy of Appropriate Empiric Antibiotic Therapy for Sepsis. Antimicrob Agents Chemother. 2010 Nov;54(11):4851–63.

13. Ferrer R, Martin-Loeches I, Phillips G, Osborn TM, Townsend S, Dellinger RP, et al. Empiric Antibiotic Treatment Reduces Mortality in Severe Sepsis and Septic Shock From the First Hour: Results From a Guideline-Based Performance Improvement Program*. Crit Care Med. 2014 Aug;42(8):1749.

14. Banerjee R, Humphries R. Rapid Antimicrobial Susceptibility Testing Methods for Blood Cultures and Their Clinical Impact. Front Med [Internet]. 2021 Mar 10 [cited 2025 Oct 21];8. Available from: https://www.frontiersin.org/journals/medicine/articles/10.3389/fmed.2021.635831/full

15. Buehler SS, Madison B, Snyder SR, Derzon JH, Cornish NE, Saubolle MA, et al. Effectiveness of Practices To Increase Timeliness of Providing Targeted Therapy for Inpatients with Bloodstream Infections: a Laboratory Medicine Best Practices Systematic Review and Meta-analysis. Clin Microbiol Rev. 2015 Nov 23;29(1):59–103.

16. Altun O, Almuhayawi M, Ullberg M, Özenci V. Clinical Evaluation of the FilmArray Blood Culture Identification Panel in Identification of Bacteria and Yeasts from Positive Blood Culture Bottles. J Clin Microbiol. 2013 Dec;51(12):4130–6.

17. 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 Oct 20;22(1):794.

18. Timbrook TT, Morton JB, McConeghy KW, Caffrey AR, Mylonakis E, LaPlante KL. The Effect of Molecular Rapid Diagnostic Testing on Clinical Outcomes in Bloodstream Infections: A Systematic Review and Meta-analysis. Clin Infect Dis. 2017 Jan 1;64(1):15–23.

19. Bauer KA, Perez KK, Forrest GN, Goff DA. Review of Rapid Diagnostic Tests Used by Antimicrobial Stewardship Programs. Clin Infect Dis. 2014 Oct 15;59(suppl_3):S134–45.

20. Lin TH, Chung HY, Jian MJ, Chang CK, Lin HH, Yu CM, et al. Artificial intelligence-clinical decision support system for enhanced infectious disease management: Accelerating ceftazidime-avibactam resistance detection in Klebsiella pneumoniae. J Infect Public Health. 2024 Oct 1;17(10):102541.

21. Jian MJ, Lin TH, Chung HY, Chang CK, Perng CL, Chang FY, et al. Artificial Intelligence-Clinical Decision Support System in Infectious Disease Control: Combatting Multidrug-Resistant Klebsiella pneumoniae with Machine Learning. Infect Drug Resist. 2024 July 10;17:2899–912.

22. Frenkel A, Rendon A, Chavez-Lencinas C, Gomez De la Torre JC, MacDermott J, Gross C, et al. Internal Validation of a Machine Learning-Based CDSS for Antimicrobial Stewardship. Life. 2025 July;15(7):1123.

23. Gomez de la Torre JC, Frenkel A, Chavez-Lencinas C, Rendon A, Cáceres JA, Alvarado L, et al. AI-Based Treatment Recommendations Enhance Speed and

Accuracy in Bacteremia Management: A Comparative Study of Molecular and Phenotypic Data. Life. 2025 June;15(6):864.

24. Beaudoin M, Kabanza F, Nault V, Valiquette L. Evaluation of a machine learning capability for a clinical decision support system to enhance antimicrobial stewardship programs. Artif Intell Med. 2016 Mar 1;68:29–36.

25. Seymour CW, Gesten F, Prescott HC, Friedrich ME, Iwashyna TJ, Phillips GS, et al. Time to Treatment and Mortality during Mandated Emergency Care for Sepsis. N Engl J Med. 2017 June 8;376(23):2235–44.

26. Lodise TP, Patel N, Kwa A, Graves J, Furuno JP, Graffunder E, et al. Predictors of 30-Day Mortality among Patients with Pseudomonas aeruginosa Bloodstream Infections: Impact of Delayed Appropriate Antibiotic Selection. Antimicrob Agents Chemother. 2007 Oct;51(10):3510–5.

27. Paterson DL, Bonomo RA. Extended-Spectrum β-Lactamases: a Clinical Update. Clin Microbiol Rev. 2005 Oct;18(4):657–86.

28. Pitout JD, Laupland KB. Extended-spectrum β-lactamase-producing Enterobacteriaceae: an emerging public-health concern. Lancet Infect Dis. 2008 Mar 1;8(3):159–66.

29. Humphries RM, Ambler J, Mitchell SL, Castanheira M, Dingle T, Hindler JA, et al. CLSI Methods Development and Standardization Working Group Best Practices for Evaluation of Antimicrobial Susceptibility Tests. J Clin Microbiol. 2018 Mar 26;56(4):10.1128/jcm.01934-17.

30. Woodford N, Turton JF, Livermore DM. Multiresistant Gram-negative bacteria: the role of high-risk clones in the dissemination of antibiotic resistance. FEMS Microbiol Rev. 2011 Sept 1;35(5):736–55.

31. MacVane SH, Nolte FS. Benefits of Adding a Rapid PCR-Based Blood Culture Identification Panel to an Established Antimicrobial Stewardship Program. J Clin Microbiol. 2016 Sept 23;54(10):2455–63.

32. Tamma PD, Cosgrove SE, Maragakis LL. Combination Therapy for Treatment of Infections with Gram-Negative Bacteria. Clin Microbiol Rev. 2012 July;25(3):450–70.

33. Magiorakos AP, Srinivasan A, Carey RB, Carmeli Y, Falagas ME, Giske CG, et al. Multidrug-resistant, extensively drug-resistant and pandrug-resistant bacteria: an international expert proposal for interim standard definitions for acquired resistance. Clin Microbiol Infect. 2012 Mar 1;18(3):268–81.

34. CLSI. Performance Standards for Antimicrobial Susceptibility Testing. 35th ed. CLSI supplement M100. Clinical and Laboratory Standards Institute; 2025.

Figure 1.- Bacteriemia Workflow program

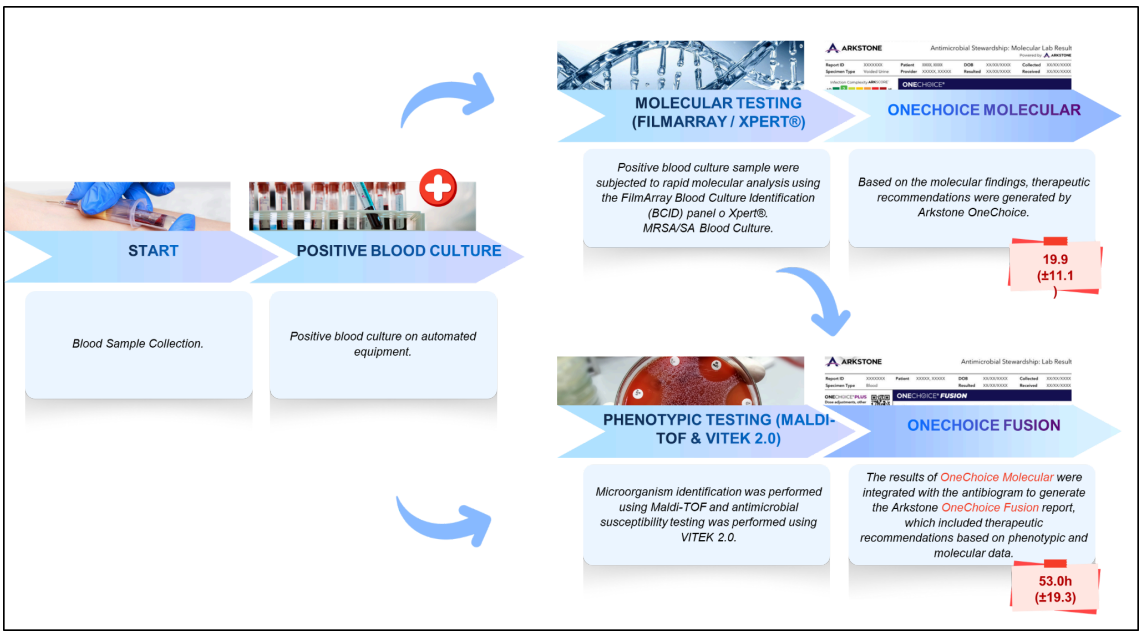

Table 1. Patient demographics and baseline characteristics

Characteristic	Value
Demographics	
Age (years), mean ± SD	58.3 ± 26.3
Age (years), median [IQR]	63 [43-78]
Male, n (%)	133 (56.1)
Female, n (%)	104 (43.9)
Clinical Characteristics	
Hospitalized, n (%)	113 (47.7)
Outpatient, n (%)	121 (51.1)
Microbiological Characteristics	
Gram-positive, n (%)	49 (20.7)
Gram-negative, n (%)	179 (75.5)
Antimicrobial resistance detected, Yes, n (%)	102 (43.0)
Antimicrobial resistance detected, No, n (%)	135 (57.0)
Most common pathogens	
Escherichia coli, n (%)	83 (35)
Klebsiella spp., n (%)	25 (10.5)
Enterococcus spp., n (%)	16 (6.8)
Enterobacter spp., n (%)	15 (6.3)
Staphylococcus aureus, n (%)	15 (6.3)

Pseudomonas aeruginosa, n (%)	14 (5.9)
Acinetobacter spp., n (%)	2 (0.8)
Diagnostic Turnaround Times	
Molecular method (hours), mean ± SD	19.7 ± 10.9
Molecular method (hours), median [IQR]	16.4 [14.1-20.6]
Conventional method (hours), mean ± SD	52.9 ± 19.4
Conventional method (hours), median [IQR]	47.8 [41.8-61.4]
Time difference (hours), mean ± SD	33.2 ± 14.2
Time difference (hours), median [IQR]	30.7 [24.6-38.9]

Fig 2:Pathogen Distribution in Bloodstream Infection Cohort (N=237)

(**A**) Horizontal bar chart showing distribution of the 10 most common pathogens: *Escherichia coli* 83 (35.0%), *Klebsiella pneumoniae* 25 (10.5%), *Salmonella* spp. 17 (7.2%), *Streptococcus* spp. 16 (6.8%), *Staphylococcus aureus* 15 (6.3%), *Enterobacter* spp. 15 (6.3%), *Pseudomonas aeruginosa* 13 (5.5%), *Enterococcus faecalis* 11 (4.6%), *Candida* spp. 8 (3.4%), *Serratia marcescens* 7 (3.0%), Other/Rare 27 (11.4%). Bars color-coded by genus/family. (**B**) Pie chart showing Gram stain classification: Gram-negative 174 (73.4%, red), Gram-positive 45 (19.0%, blue), Fungi 8 (3.4%, green), Not detected/Outside panel 8 (3.4%, gray). Predominance of Gram-negative bacteria and *E. coli* reflects typical BSI epidemiology with additional burden of enteric pathogens (*Salmonella*) in Peru.

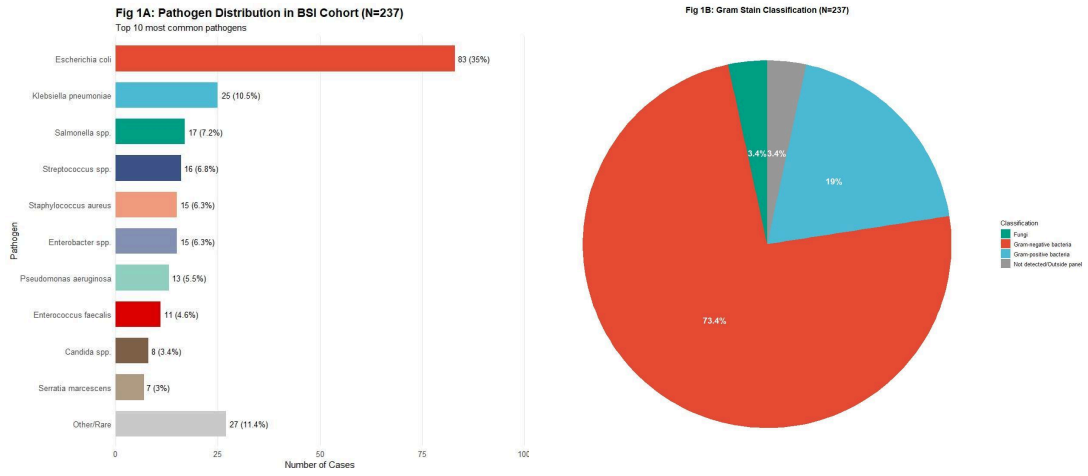

Fig 3: Distribution of Diagnostic Turnaround Times: Molecular vs Conventional Methods (N=237)

Histogram panels showing frequency distribution of turnaround times (hours from blood draw to result availability) for (**A**) molecular diagnostics (FilmArray BCID or Xpert MRSA/SA BC), mean 19.7±10.9 hours, median 16.4 hours (blue); (**B**) conventional culture with MALDI-TOF identification and phenotypic antimicrobial susceptibility testing, mean 52.9±19.4 hours, median 47.5 hours (red); and (**C**) time saved by molecular methods (conventional minus molecular turnaround time), mean 33.2±14.2 hours, median 30.7 hours, representing 62.7% time reduction (purple). Dashed red line = mean, dashed blue line = median. Wilcoxon signed-rank test $P<0.001$, Cohen's $d=2.33$ (very large effect size).

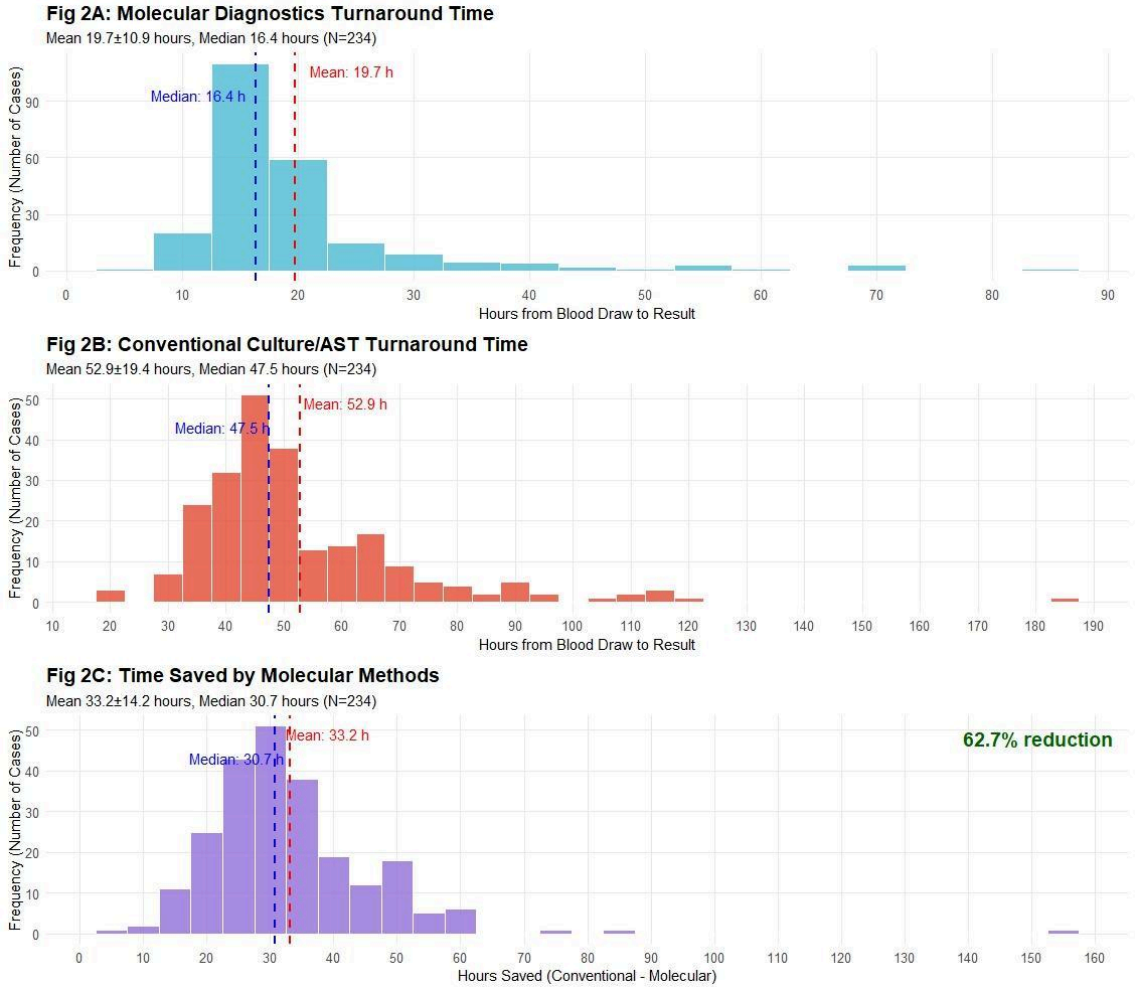

Fig 4:Cumulative Distribution Functions: Molecular vs Conventional Diagnostic Methods (N=237)

Cumulative probability plots showing proportion of cases achieving diagnostic results as a function of time. Green curve = molecular diagnostics (FilmArray/Xpert), median 16.4 hours. Red curve = conventional culture/phenotypic AST, median 47.5 hours. Horizontal dashed line indicates 50th percentile (median). Vertical dashed lines mark respective medians. The rightward shift of the conventional curve relative to molecular curve illustrates substantial time delay. At 20 hours, ~60% of molecular diagnostics are

complete vs ~5% of conventional methods. At 48 hours, 95% molecular complete vs ~55% conventional. Wilcoxon signed-rank test *P*<0.001.

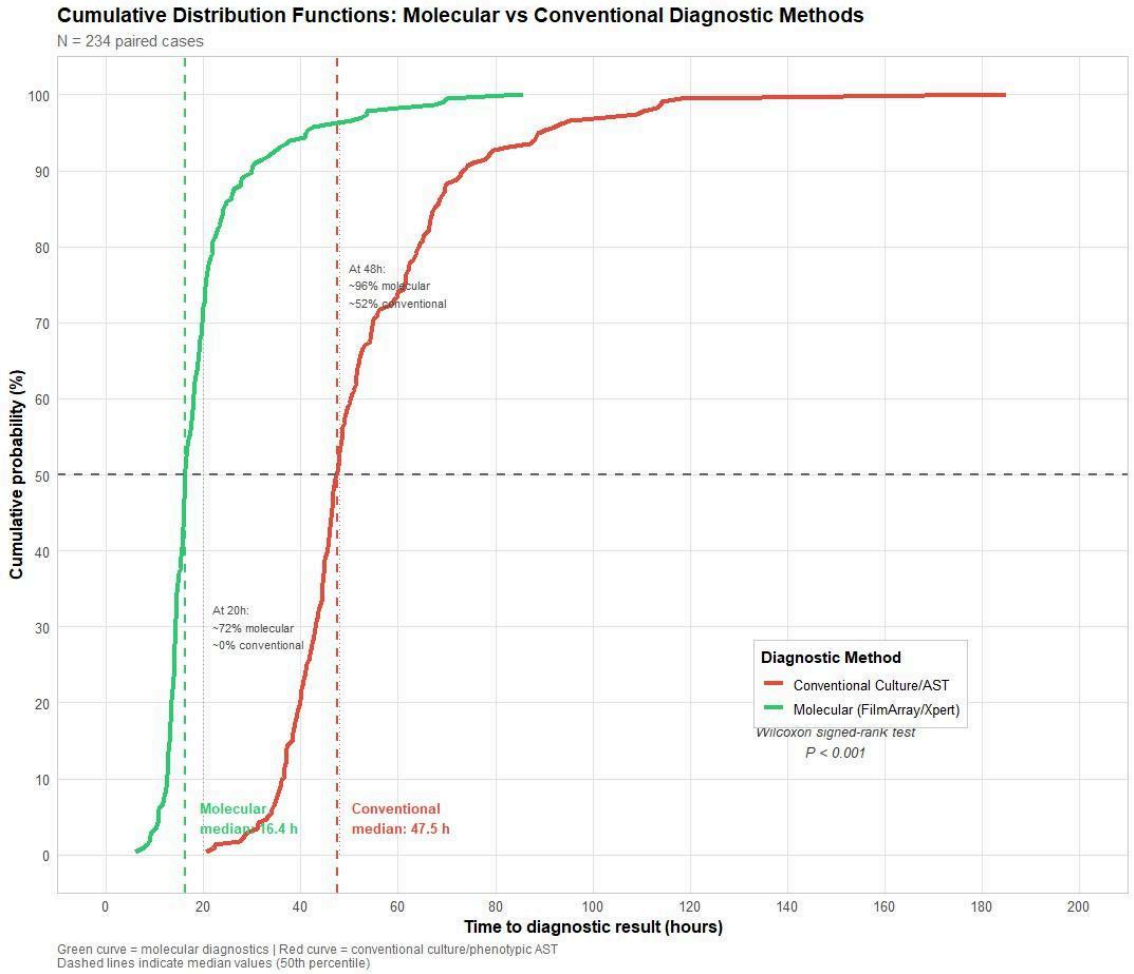

Table 2:Diagnostic Turnaround Time by Clinical Characteristics

Characteristic	N	Molecular Mean ± SD	(h)Phenotypic Mean ± SD	(h)Difference Mean ± SD	(h)P-value
GLOBAL	237	19.9 ± 11.1	53.0 ± 19.3	33.1 ± 14.2	<0.001†

By Gram classification					
Gram-negative	179	19.4 ± 11.4	50.0 ± 16.8	30.6 ± 11.1	<0.001†
Gram-positive	49	18.8 ± 5.9	58.1 ± 13.1	39.3 ± 10.6	<0.001†
Fungi	8	37.3 ± 16.0	92.7 ± 43.1	55.3 ± 42.4	0.008†
By resistance presence					
With resistance	102	17.9 ± 10.2	48.7 ± 16.0	30.8 ± 11.6	<0.001†
Without resistance	135	21.4 ± 11.6	56.2 ± 20.9	34.8 ± 15.7	<0.001†
By clinical setting					
Hospitalized	113	19.4 ± 11.1	52.8 ± 18.1	33.4 ± 12.8	<0.001†
Ambulatory	121	20.5 ± 11.3	53.1 ± 20.6	32.7 ± 15.4	<0.001†
By specific pathogen					
Escherichia coli	82	18.3 ± 11.3	46.3 ± 14.3	28.0 ± 8.1	<0.001†
Klebsiella pneumoniae	23	16.1 ± 4.7	47.3 ± 15.7	31.3 ± 14.9	<0.001†
Staphylococcus aureus	13	16.4 ± 2.3	50.0 ± 8.0	33.6 ± 6.9	<0.001†
Pseudomonas aeruginosa	14	22.7 ± 13.7	55.6 ± 15.0	32.9 ± 12.8	<0.001†
Salmonella spp.	17	19.6 ± 5.1	47.0 ± 8.5	27.4 ± 8.9	<0.001†

Notes:
† Wilcoxon paired test comparing molecular vs. phenotypic times within each subgroup
SD=Standard Deviation
h = hours

TABLE 2A: Bacterial Identification Concordance By Clinical Subgroups

Clinical_Characteristic	N	Complete n (%)	Partial n (%)	Discordant n (%)	Combined %	95% CI
OVERALL	237	189 (79.7%)	35 (14.8%)	13 (5.5%)	94.5%	(90.8-96.8%)

By Gram Classification						
Gram-negative bacteria	179	149 (83.2%)	22 (12.3%)	8 (4.5%)	95.5%	(91.4-97.7%)
Gram-positive bacteria	49	32 (65.3%)	13 (26.5%)	4 (8.2%)	91.8%	(80.8-96.8%)
Fungi	8	8 (100.0%)	0 (0.0%)	0 (0.0%)	100.0%	(67.6-100.0%)
By Resistance Status						
With antimicrobial resistance	102	92 (90.2%)	7 (6.9%)	3 (2.9%)	97.1%	(91.7-99.0%)
Without resistance	135	97 (71.9%)	28 (20.7%)	10 (7.4%)	92.6%	(86.9-95.9%)
By Clinical Setting						
Hospitalized patients	113	100 (88.5%)	9 (8.0%)	4 (3.5%)	96.5%	(91.3-98.6%)
Outpatient/Community	121	87 (71.9%)	25 (20.7%)	9 (7.4%)	92.6%	(86.5-96.0%)
By Specific Pathogen						
Escherichia coli	83	82 (98.8%)	0 (0.0%)	1 (1.2%)	98.8%	(93.4-99.8%)
Klebsiella pneumoniae	23	20 (87.0%)	2 (8.7%)	1 (4.3%)	95.7%	(79.0-99.2%)
Staphylococcus aureus	13	13 (100.0%)	0 (0.0%)	0 (0.0%)	100.0%	(77.2-100.0%)
Salmonella spp.	17	2 (11.8%)	15 (88.2%)	0 (0.0%)	100.0%	(81.6-100.0%)
Streptococcus spp.	16	5 (31.2%)	11 (68.8%)	0 (0.0%)	100.0%	(80.6-100.0%)

Complete: Molecular and MALDI-TOF identify same species
Partial: Molecular identifies genus, MALDI-TOF identifies genus + species (clinically valuable)
Discord: Methods disagree (mainly pathogens outside molecular panel)
Combined: Complete + Partial concordance

TABLE 2B: Antimicrobial Resistance Concordance By Clinical Subgroups

Clinical_Characteristic	N	Complete n (%)	Partial n (%)	Discordant (%)	nCombined %	95% CI
OVERALL	237	210 (88.6%)	2 (0.8%)	25 (10.5%)	89.5%	(84.9-92.8%)
By Gram Classification						
Gram-negative bacteria	179	162 (90.5%)	2 (1.1%)	15 (8.4%)	91.6%	(86.6-94.9%)
Gram-positive bacteria	49	41 (83.7%)	0 (0.0%)	8 (16.3%)	83.7%	(71.0-91.5%)
Fungi	8	7 (87.5%)	0 (0.0%)	1 (12.5%)	87.5%	(52.9-97.8%)
By Resistance Status						
With antimicrobial resistance	102	88 (86.3%)	2 (2.0%)	12 (11.8%)	88.2%	(80.6-93.1%)
Without resistance	135	122 (90.4%)	0 (0.0%)	13 (9.6%)	90.4%	(84.2-94.3%)
By Clinical Setting						
Hospitalized patients	113	99 (87.6%)	2 (1.8%)	12 (10.6%)	89.4%	(82.4-93.8%)
Outpatient/Community	121	108 (89.3%)	0 (0.0%)	13 (10.7%)	89.3%	(82.5-93.6%)
By Specific Pathogen						
Escherichia coli	82	78 (95.1%)	2 (2.4%)	2 (2.4%)	97.6%	(91.5-99.3%)
Klebsiella pneumoniae	23	22 (95.7%)	0 (0.0%)	1 (4.3%)	95.7%	(79.0-99.2%)
Staphylococcus aureus	13	13 (100.0%)	0 (0.0%)	0 (0.0%)	100.0%	(77.2-100.0%)
Salmonella spp.	17	16 (94.1%)	0 (0.0%)	1 (5.9%)	94.1%	(73.0-99.0%)
Streptococcus spp.	16	11 (68.8%)	0 (0.0%)	5 (31.2%)	68.8%	(44.4-85.8%)

Complete: Genotype and phenotype agree on resistance mechanism
Partial: Minor discrepancy in resistance detection
Discord: Gene+/Phenotype- or Gene-/Phenotype+ (mainly chromosomal AmpC)
Combined: Complete + Partial concordance

Fig 5:Timeline Comparison of Diagnostic-Therapeutic Strategies

Horizontal timeline comparing three diagnostic-therapeutic strategies from blood culture draw (0 hours) to therapy decision point. (**Strategy 1**: Conventional Only) Single red bar extending to 52.9 hours for conventional culture/AST completion before therapy decision. (**Strategy 2**: Molecular Only) Single green bar extending to 19.7 hours for molecular diagnostics/OneChoice AI, with annotation showing 81.4% concordance rate and orange arrow indicating 33.2-hour time advantage vs

conventional. (**Strategy 3**: Integrated/Recommended) Blue bar to 19.7 hours (molecular/OneChoice initial therapy) followed by purple bar extending to 52.9 hours (conventional/Fusion optimization), with dual decision points marked. Clinical outcomes annotation box: 81.4% receive appropriate therapy without adjustment (green), 18.6% receive optimization (purple), 100% receive appropriate therapy ultimately. Time saved annotation emphasizes 33.2-hour therapeutic window representing 62.7% reduction.

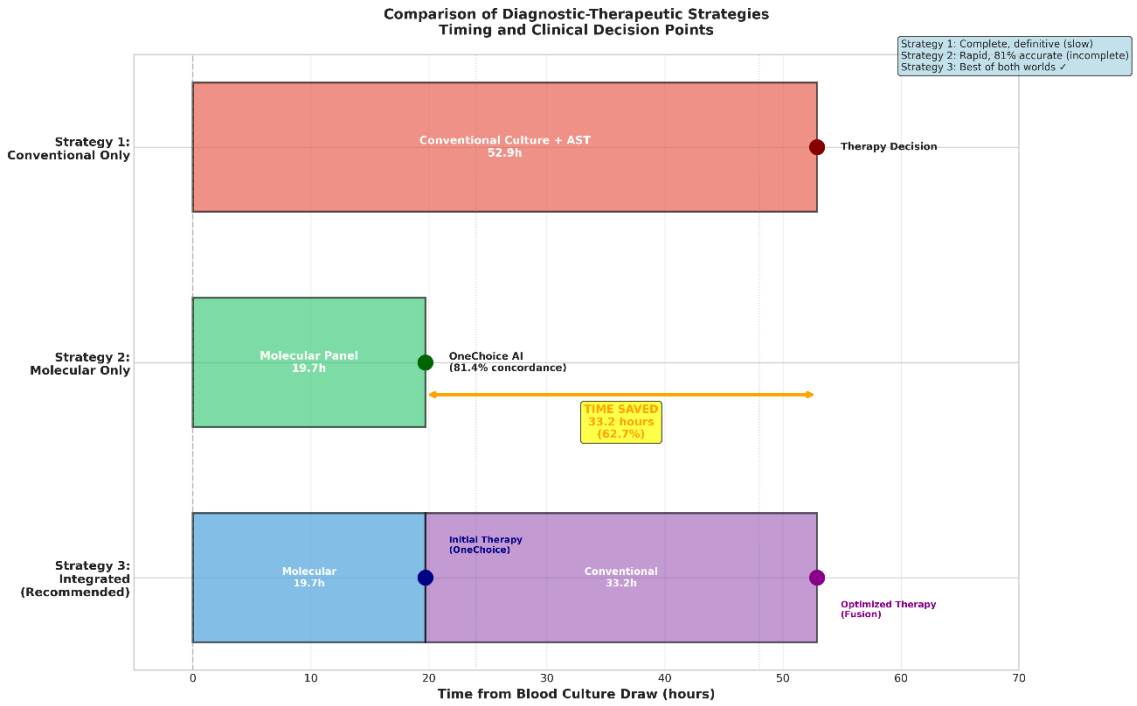

TABLE 4: Therapeutic Concordance By Clinical Subgroups

Clinical Characteristic	N	Concordant n(%)	Discordant n(%)	Concordance %	95% CI
OVERALL	236	192 (81.4%)	44 (18.6%)	81.4%	(75.9-85.8%)

By Gram Classification					
Gram-negative bacteria	179	151 (84.4%)	28 (15.6%)	84.4%	(78.3-89.0%)
Gram-positive bacteria	48	34 (70.8%)	14 (29.2%)	70.8%	(56.8-81.8%)
By Resistance Status					
With antimicrobial resistance	102	87 (85.3%)	15 (14.7%)	85.3%	(77.1-90.9%)
Without resistance	134	105 (78.4%)	29 (21.6%)	78.4%	(70.6-84.5%)
By Bacterial ID Concordance					
Bacterial ID concordant	189	163 (86.2%)	26 (13.8%)	86.2%	(80.6-90.4%)
Bacterial ID discordant	47	29 (61.7%)	18 (38.3%)	61.7%	(47.4-74.2%)
By Resistance Detection Concordance					
Resistance detection concordant	209	184 (88.0%)	25 (12.0%)	88.0%	(82.9-91.8%)
Resistance detection discordant	27	8 (29.6%)	19 (70.4%)	29.6%	(15.9-48.5%)
By Clinical Setting					
Hospitalized patients	112	90 (80.4%)	22 (19.6%)	80.4%	(72.0-86.7%)
Outpatient/Community	121	99 (81.8%)	22 (18.2%)	81.8%	(74.0-87.7%)
By Specific Pathogen					
Escherichia coli	82	78 (95.1%)	4 (4.9%)	95.1%	(88.1-98.1%)
Klebsiella pneumoniae	23	21 (91.3%)	2 (8.7%)	91.3%	(73.2-97.6%)
Staphylococcus aureus	13	13 (100.0%)	0 (0.0%)	100.0%	(77.2-100.0%)

Concordant: OneChoice (molecular-based) and OneChoice Fusion (molecular+phenotypic) recommend same antibiotic
Discordant: Recommendations differ due to additional phenotypic susceptibility data

TABLE 5: Predictors Of Therapeutic Discordance

Predictor	Univariate OR	Univariate P-value	Adjusted OR	95% CI	Adjusted P-value
Resistance Detection Discordant	9.16	<0.001***	7.70	3.61 - 17.49	<0.001***
Gram-Positive (vs Gram-Negative)	2.10	0.031*	1.77	0.84 - 3.56	0.124
Hospitalized (vs Outpatient)	1.13	0.701	1.24	0.65 - 2.30	0.516
Resistance Present (vs None)	0.64	0.156	0.69	0.33 - 1.45	0.316
Bacterial ID Discordant	6.31	<0.001***	2.76	1.20 - 6.32	0.019*
Male (vs Female)	0.76	0.369	0.76	0.42 - 1.51	0.395
Age (per year)	1.00	0.858	1.00	0.99 - 1.02	0.501
Molecular Turnaround Time	1.02	0.117	1.02	0.99 - 1.06	0.197

OR: Odds Ratio; CI: Confidence Interval. Multivariable logistic regression analysis was performed to identify independent predictors of therapeutic discordance, defined as disagreement between OneChoice and OneChoice Fusion treatment recommendations. All variables shown were included simultaneously in the multivariable model (N = 224 complete cases, 42 discordant cases [18.8%]). Model fit: Pseudo R^2 (McFadden) = 0.202; Likelihood Ratio Test: $\chi^2 = 43.59$, df = 8, $P < 0.001$. * $P < 0.05$; ** $P < 0.01$; *** $P < 0.001$.