ORIGINAL RESEARCH PAPER

Artificial Intelligence in Antimicrobial Susceptibility Testing: Transforming Clinical Microbiology from Plates to Prediction

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Abstract

Background: Antimicrobial susceptibility testing (AST) is central to clinical microbiology and antimicrobial stewardship. Conventional AST methods, though standardized, are time-consuming and subject to inter-observer variability. Artificial intelligence (AI) has emerged as a potential tool to enhance standardization and efficiency in AST interpretation.

Objectives: This study evaluated the performance of AI-assisted interpretation of disk diffusion AST compared with conventional manual interpretation in a routine clinical microbiology laboratory.

Methods: A laboratory based retrospective analytical study was conducted on 300 non-duplicate bacterial isolates. AST was performed using Kirby–Bauer disk diffusion following CLSI guidelines. High-resolution images of AST plates were analysed using an AI-assisted image interpretation system. Results were compared with manual readings by experienced microbiologists. Categorical agreement, error rates, and interpretation time were assessed.

Results: Overall categorical agreement between AI-assisted and manual interpretation was 96.2%. Very major errors, major errors, and minor errors were 1.2%, 1.5%, and 3.8%, respectively. AI-assisted interpretation significantly reduced interpretation time while maintaining acceptable accuracy.

Conclusion: AI-assisted AST interpretation can serve as a reliable adjunct to conventional methods, improving efficiency and consistency while preserving the essential role of microbiologist oversight.

Keywords: Artificial intelligence, antimicrobial susceptibility testing, disk diffusion, clinical microbiology, automation, antimicrobial resistance

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Introduction

Antimicrobial resistance (AMR) is a growing global health crisis that threatens the effective treatment of infectious diseases. Antimicrobial susceptibility testing (AST) plays a pivotal role in guiding appropriate antimicrobial therapy, preventing misuse of antibiotics, and supporting antimicrobial stewardship programs. Despite advances in diagnostic technologies, phenotypic AST methods such as disk diffusion and minimum inhibitory concentration (MIC) determination remain the backbone of susceptibility testing in most clinical microbiology laboratories.

Disk diffusion AST is cost-effective, standardized, and widely applicable, particularly in resource-limited settings. However, it requires meticulous manual measurement of inhibition zones and interpretation based on breakpoint criteria. These processes are labor-intensive and subject to inter- and intra-observer variability, especially in borderline results near interpretative breakpoints.

Laboratory workloads have increased substantially due to rising sample volumes and the emergence of multidrug-resistant organisms. Concurrently, shortages of trained microbiology personnel have intensified operational pressures. While automated susceptibility systems address some of these challenges, their high cost and infrastructural requirements limit universal adoption.

Artificial intelligence (AI), particularly image analysis and machine learning techniques, offers a novel approach to addressing these limitations. By automating zone measurement and interpretation, AI-assisted AST has the potential to enhance standardization, reduce human error, and improve turnaround time. This study assesses the performance and feasibility of AI-assisted AST interpretation in comparison with conventional manual reading in a clinical microbiology laboratory.

Materials and Methods

Study Design and Setting

A laboratory based retrospective analytical study was conducted over six months in the Department of Clinical Microbiology, Government Ramanathapuram Medical College Hospital, Tamil Nadu, India. The study utilized anonymized routine antimicrobial susceptibility testing data generated as part of standard patient care.

Ethical Considerations

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This study was based exclusively on anonymized antimicrobial susceptibility testing data generated during routine clinical diagnostic services. No patient identifiers were accessed, and no additional samples were collected for research purposes. As the analysis involved secondary laboratory data without direct patient interaction or intervention, it did not require formal institutional ethical review.

Bacterial Isolates

A total of 300 non-duplicate bacterial isolates obtained from clinical specimens such as blood, urine, pus, respiratory samples, and body fluids were included. Both Gram-positive and Gram-negative organisms commonly encountered in clinical practice were analysed. A convenience sample of 300 consecutive non-duplicate isolates was included during the study period to ensure adequate representation of commonly encountered pathogens.

Distribution of Isolates

Table 1. Distribution of Bacterial Isolates Included in the Study (n = 300)

Organism	**Number of Isolates (%)**
Escherichia coli	90 (30.0)
Klebsiella pneumoniae	60 (20.0)
Pseudomonas aeruginosa	45 (15.0)
Staphylococcus aureus	55 (18.3)
Enterococcus spp.	30 (10.0)
Other Gram-negative bacilli	20 (6.7)
Total	**300 (100)**

Antimicrobial Susceptibility Testing

AST was performed using the Kirby–Bauer disk diffusion method on Mueller–Hinton agar following Clinical and Laboratory Standards Institute (CLSI) guidelines. Antibiotic panels were selected based on organism type and institutional antibiotic policy. Plates were incubated at 35–37°C for 16–18 hours under aerobic conditions.

Manual Interpretation

Zone diameters were measured manually using a calibrated ruler by two experienced microbiologists blinded to AI-generated results. Discrepancies were resolved by consensus. Interpretations were categorized as susceptible, intermediate, or resistant using CLSI breakpoints.

AI-Assisted Interpretation

AST plates were photographed using a standardized imaging setup to ensure uniform lighting and distance. AST plate images were analysed using a deep learning based image analysis algorithm trained to detect inhibition zones and classify susceptibility categories based on CLSI breakpoints. The AI system functioned as a decision-support tool and was not modified during the study period. The algorithm utilized convolutional neural network (CNN)-based image segmentation for zone detection and rule-based classification according to CLSI breakpoint thresholds. Internal validation was performed using a subset of laboratory images prior to implementation. The algorithm was applied in a blinded manner, and its outputs were compared independently with manual readings. The AI system was evaluated for performance and was not used for clinical decision-making during the study period.

Outcome Measures

Primary and secondary outcomes included:

- Categorical agreement between AI and manual interpretation
- Major, very major, and minor error rates
- Time required for interpretation per plate

Statistical Analysis

Statistical analysis was performed using IBM SPSS Statistics (Version 31.0). Categorical agreement was calculated as the percentage concordance between manual and AI-assisted interpretation. Cohen's kappa coefficient was used to assess agreement beyond chance. Error rates were calculated according to CLSI performance evaluation criteria.

Results

The overall Cohen's kappa coefficient between manual and AI-assisted interpretation was 0.92, indicating excellent agreement. The kappa value of 0.92 indicates excellent agreement according to Landis and Koch interpretation criteria.

Agreement Between Manual and AI-Assisted Interpretation

Overall categorical agreement between AI-assisted and manual AST interpretation was 96.2%. Most discrepancies occurred in isolates with zone diameters close to interpretative breakpoints.

Table 2. Agreement and Error Rates Between AI and Manual AST Interpretation

Parameter	**Value (%)**
Categorical agreement	96.2
Very major errors (false susceptible)	1.2
Major errors (false resistant)	1.5
Minor errors	3.8

Error rates were within acceptable limits recommended for AST performance evaluation.

Time Efficiency and Workflow Comparison

AI-assisted interpretation substantially reduced the time required for AST reading.

Table 3. Comparison of Manual and AI-Assisted AST Interpretation

Parameter	**Manual Interpretation**	**AI-Assisted Interpretation**
Mean interpretation time per plate	90–120 seconds	15–20 seconds

Inter-observer variability	Present	Not applicable
Standardization	Operator-dependent	Algorithm-based
Fatigue-related variation	Possible	Absent
Requirement for expert oversight	Required	Required (adjunct role)

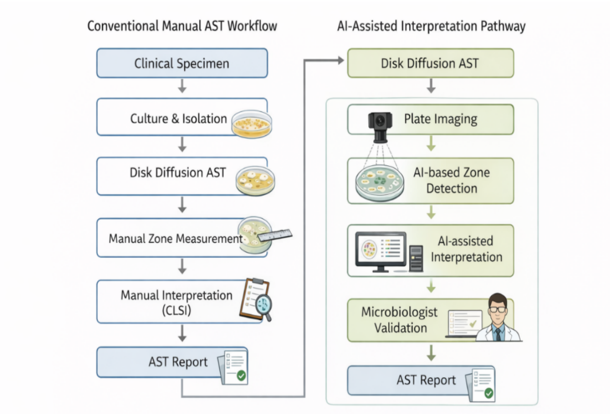


Figure 1. Flowchart illustrating conventional manual antimicrobial susceptibility testing workflow and AI-assisted AST interpretation pathway.

Figure 2. Concordance Between Manual and AI AST Interpretation

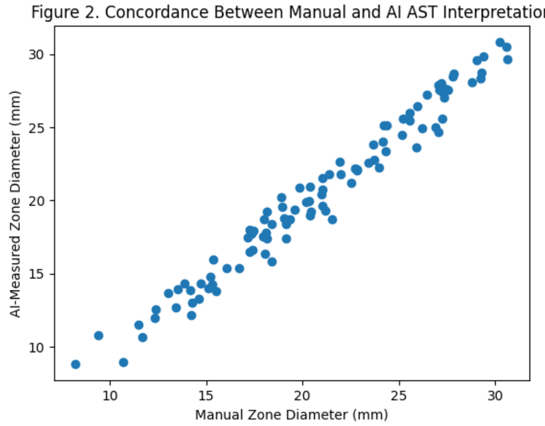
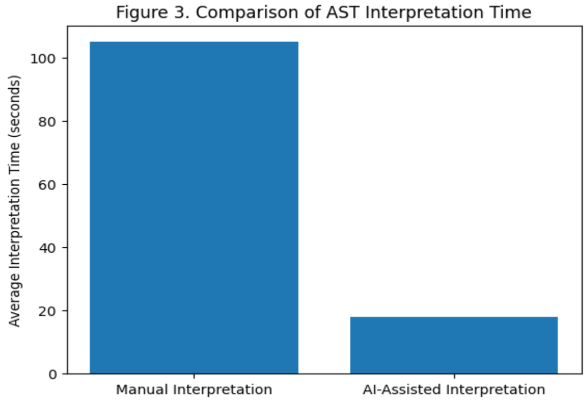


Figure 3. Comparison of AST Interpretation Time



Discussion

This study demonstrates that AI-assisted interpretation of disk diffusion AST achieves high concordance with conventional manual readings while offering significant improvements in efficiency and consistency. The high categorical agreement observed supports the feasibility of integrating AI tools into routine microbiology workflows.
Inter-observer variability is a recognized limitation of manual AST interpretation, particularly in high-volume laboratories and borderline cases. AI-assisted systems apply uniform interpretative criteria, thereby enhancing reproducibility and reducing subjective variation. The marked reduction in interpretation time observed in this study may help laboratories manage increasing workloads without compromising quality.
Importantly, AI-assisted AST should not be viewed as a replacement for microbiologists. Human expertise remains essential for recognizing technical anomalies, unusual resistance patterns, and clinically discordant results. AI functions best as a supportive tool that augments, rather than replaces, professional judgment.
The potential impact of AI-assisted AST is particularly relevant in resource-limited settings, where access to automated susceptibility platforms may be restricted. Image-based AI systems combined with conventional disk diffusion offer a scalable and cost-effective alternative.

Limitations

The study was conducted at a single center and did not include molecular resistance mechanisms or clinical outcome correlation. Rare pathogens and emerging resistance patterns were underrepresented. The AI algorithm was evaluated using internally generated laboratory data and did not undergo external validation. Further multicenter external validation studies are required before routine clinical implementation.

Conclusion

Artificial intelligence–assisted antimicrobial susceptibility testing represents a promising adjunct to conventional AST methods. By improving consistency and reducing interpretation time, AI can strengthen microbiology laboratory workflows while preserving the central role of microbiologist expertise. Careful validation, quality control, and ethical integration are essential for successful implementation.

Ethical Statement and AI Use Disclosure

The AI-based image analysis system described in this study was evaluated as a laboratory decision-support tool. The authors independently verified all results and interpretations.

References

1. World Health Organization. *Global action plan on antimicrobial resistance.* WHO; 2015.
2. Clinical and Laboratory Standards Institute. *Performance Standards for Antimicrobial Susceptibility Testing (CLSI M100).* Latest edition.
3. van Belkum A, et al. Innovations in antimicrobial susceptibility testing. *Nat Rev Microbiol.* 2020;18:299–311.

4. Bostrom E, et al. Artificial intelligence-based analysis of disk diffusion susceptibility testing. *J Clin Microbiol.* 2022;60:e02198-21.
5. Smith KP, Kirby JE. Image analysis and automation in clinical microbiology. *Clin Microbiol Rev.* 2020;33:e00062-19.
6. Burnham CAD, Leeds J, Nordmann P, et al. Diagnosing antimicrobial resistance. *Nat Rev Microbiol.* 2017;15:697–703.
7. Humphries RM, et al. The role of susceptibility testing in antimicrobial stewardship. *Clin Infect Dis.* 2018;67:512–518.
8. Taha M, et al. Machine learning applications in antimicrobial resistance prediction. *Front Microbiol.* 2021;12:657890.
9. EUCAST. *Routine and extended internal quality control for antimicrobial susceptibility testing.*
10. O'Neill J. *Tackling drug-resistant infections globally.* Review on Antimicrobial Resistance; 2016.
11.