TRENDS AND DEVELOPMENTS IN MOLECULAR DIAGNOSTICS FOR ANTIBIOTIC RESISTANCE DETECTION

Sintya Agy Nuraisah Isang
Bina Mandiri University , Gorontalo,
E-mail: sintyagyn@students.ubmg.ac.id

ABSTRACT

Antimicrobial Resistance (AMR) poses a global threat, necessitating a shift from slow culture-based methods (24–48 hours) to rapid molecular diagnostics to ensure timely targeted therapy. Methodology: This study employed a Systematic Literature Review (SLR) and bibliometric analysis of 10,776 Scopus-indexed articles (up to 2024) using the PRISMA protocol and VOSviewer for network visualization. Publications peaked in 2024, with the United States and China leading global contributions. Thematic analysis reveals a transition from basic genetic studies to advanced technologies like NGS, multiplex PCR, CRISPR-based systems, and microfluidic biosensors. Key markers include carbapenemase (bla_{NDM}, bla_{KPC}), colistin resistance (mcr-1), and ESBLs. Emerging trends emphasize AI integration and decentralized Point-of-Care (POC) testing. Conclusion: Molecular diagnostics are vital for Antimicrobial Stewardship. Despite technological leaps, clinical standardization and costs remain significant barriers. This mapping provides a strategic roadmap for enhancing global resistance surveillance and precision diagnostics.

Keywords : Antimicrobial resistance; Molecular diagnostics; PCR; Next-generation sequencing; CRISPR-Cas; Point-of-care testing.

INTRODUCTION

The global crisis of antimicrobial resistance (AMR) has reached a critical juncture, fundamentally compromising the efficacy of modern medicine and returning humanity to an era where common infections could once again become fatal. As pathogenic bacteria evolve diverse defense mechanisms against existing drug classes, the clinical utility of standard antibiotic treatments continues to diminish, leading to prolonged hospital stays, increased economic burdens on healthcare systems, and significantly higher mortality rates worldwide. The World Health Organization has identified AMR as one of the top global public health threats, emphasizing that without urgent intervention, the "post-antibiotic era" could become a devastating reality.

Historically, the management of infectious diseases has relied heavily on traditional culture-based diagnostic methods. While these phenotypic approaches remain the "gold standard" for determining actual bacterial susceptibility, they are inherently limited by their slow turnaround times, typically requiring 24 to 48 hours or even longer for fastidious organisms to yield actionable results. This diagnostic lag often forces clinicians to initiate broad-spectrum empirical therapy, which may be suboptimal or entirely ineffective against resistant strains. Such imprecise prescribing not only jeopardizes patient outcomes but also creates selective pressure that further accelerates the emergence and dissemination of multi-drug resistant organisms (MDROs). Consequently, there is a profound and urgent clinical necessity for the development and implementation of rapid, highly sensitive, and specific diagnostic tools that can guide targeted therapy within

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the "golden hour" of infection management.

Molecular diagnostics have revolutionized this paradigm by shifting the focus from observing bacterial growth to the direct detection of resistance-conferring genetic determinants. Over the past two decades, the field has transitioned from basic Polymerase Chain Reaction (PCR) assays to a sophisticated suite of advanced technologies. Next-Generation Sequencing (NGS) now enables comprehensive whole-genome profiling of pathogens, while innovative platforms such as CRISPR-based diagnostics and Loop-mediated Isothermal Amplification (LAMP) offer unprecedented speed and portability. These molecular methodologies allow for the precise identification of high-priority markers, such as carbapenemases (*blaNDM*, *blaKPC*), colistin resistance genes (*mcr-1*), and extended-spectrum β-lactamases (ESBLs), often delivering results in a fraction of the time required by conventional methods.

Despite the exponential proliferation of scientific literature in this domain, the sheer volume of data makes it challenging for researchers and policymakers to discern the overarching intellectual structure and shifting research priorities. A comprehensive visualization of the global research landscape is essential to identify established knowledge bases, emerging research frontiers, and existing gaps in the literature. Therefore, this study utilizes bibliometric analysis and science mapping techniques to systematically evaluate the evolution of molecular diagnostics for antibiotic resistance. By highlighting the most influential authors, key institutional collaborations, and emerging technological clusters, this research provides a strategic roadmap for the future of clinical microbiology and the global fight against antimicrobial resistance.

RESEARCH METHODS

A Systematic Literature Review (SLR) combined with a bibliometric approach was employed to quantitatively assess the literature, identifying trends, patterns, and key research actors within the discipline of Medical Laboratory Technology, specifically in the field of molecular diagnostics. By applying established frameworks such as PRISMA (Preferred Reporting Items for Systematic Reviews and Meta-Analyses), this methodology ensures a comprehensive and replicable assessment of the literature, producing a clear and transparent overview of the research topic. The PRISMA flow diagram guarantees transparency and completeness in the review process, allowing readers to understand the core procedures used and to examine the reduction (or filtering) of irrelevant records [1][2]. The established inclusion criteria were: (1) articles published up to December 31, 2024, (2) publications in English, and (3) a focus on the theme of "Trends and Developments in Molecular Diagnostics for Antibiotic Resistance Detection."

Bibliometric analysis is a powerful tool for assessing the impact and trends in scientific research. By systematically analyzing large volumes of bibliographic data, this analysis provides valuable insights that can guide research directions and technological developments in medical laboratories [3][4][5]. The bibliometric analysis was conducted using software such as VOSviewer to visualize bibliographic data, including citation networks, author collaborations, and keyword co-occurrences, thereby revealing the intellectual structure and dynamics within the field of study. The integration of both approaches SLR and Bibliometrics offers a holistic understanding of the evolution, historical trajectory, and future directions of the diagnostic research domain, making it beneficial in clinical studies for deeper insights.

The initial phase of this academic study began with keyword selection using a macro (top-down) approach. The primary focus was set on the use of keywords *("molecular diagnostic" OR "PCR" OR "NGS") AND ("antibiotic resistance" OR "AMR")** within the title, abstract, and keywords of the articles. Subsequently, the Scopus database was utilized by the researcher to conduct an extensive literature review, identify experts in the field of molecular microbiology, and monitor current trends in diagnostic technology.

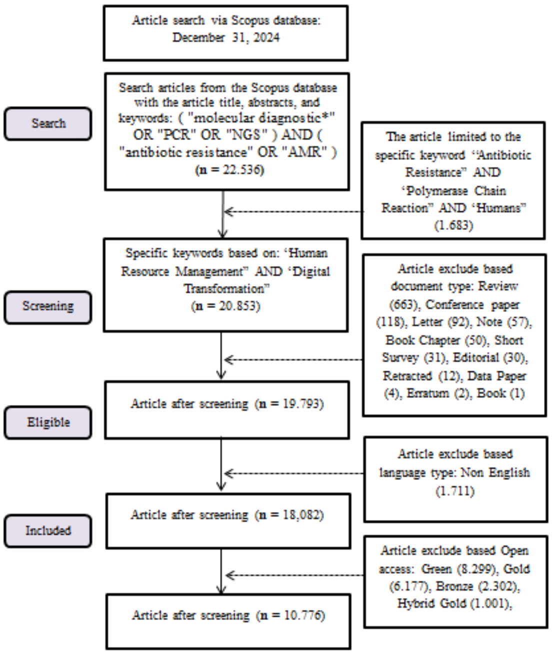


Figure 1. PRISMA Flow Diagram for Systematic Literature Review Information

Based on the initial search results retrieved from the Scopus database on December 31, 2024, a total of 22,536 documents related to molecular diagnostics and antibiotic resistance were identified (See Figure 1). The search was further narrowed using specific keywords such as *"Antibiotic Resistance"*, *"Polymerase Chain Reaction"*, and *"Humans"*, which filtered the documents down to 1,683 potential records. However, to maintain relevance to the management and digital transformation aspects within the laboratory, additional filtering based on the keywords *"Human Resource Management"* and *"Digital Transformation"* was performed, resulting in 20,853 related documents within a broader spectrum.

The identified documents then underwent a rigorous screening process. The first stage excluded documents based on publication type: Reviews (663), Conference Papers (118), Letters (92), Notes (57), Book Chapters (50), Short Surveys (31), Editorials (30), Retracted (12), Data Papers (4), Errata (2), and Books (1), leaving 19,793 documents. Further refinement was achieved by excluding 1,711 articles not written in English, leaving 18,082 eligible articles. The final screening step involved exclusion based on accessibility status, where document categories such as Green (8,299), Gold (6,177), Bronze (2,302), and Hybrid Gold (1,001) were processed, resulting in a final analysis set of 10,776 documents. These 10,776 final articles were then subjected to in-depth analysis in this study to answer the following Research Questions (RQ):

a. RQ1: What are the primary conceptual or thematic clusters, emerging technologies (*research frontiers*), and trends in the literature regarding Molecular Diagnostics for Antibiotic Resistance Detection?
b. RQ2: Who are the key authors, institutions, and countries contributing to the most significant research in this field, and what are the patterns of their global research collaboration networks?
c. RQ3: What are the specific molecular markers, diagnostic methodologies, and key research findings highlighted in the most influential literature regarding antibiotic resistance detection?

RESEARCH RESULT

The findings of this study are based on 10,776 articles identified from the Scopus database, specifically addressing Trends and Developments in Molecular

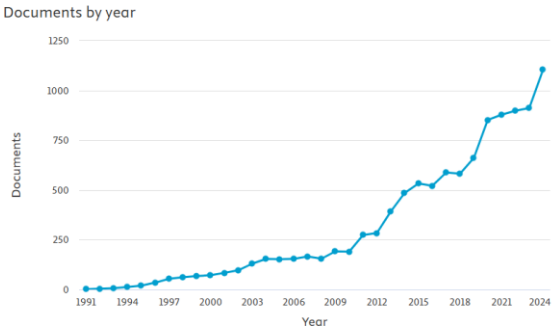
Diagnostics for Antibiotic Resistance Detection. Data derived from these articles were meticulously analyzed by examining the total volume of publications, annual publication trends, and key journal sources within the fields of clinical microbiology and medical laboratory technology. Furthermore, this study highlights the most influential elements in the literature on this topic, including contributing authors in the development of detection techniques (such as PCR and NGS), their institutional affiliations, and the international collaboration networks involved in global antimicrobial resistance mapping. Through the integration of bibliometric analysis and systematic literature review, this study also dissects the most effective diagnostic methodologies and the genetic markers (resistance genes) most frequently reported in the indexed scientific literature.

DISCUSSION

RQ1: What are the primary conceptual or thematic clusters, emerging technologies (*research frontiers*), and trends in the literature regarding Molecular Diagnostics for Antibiotic Resistance Detection?

Bibliometric analysis of 10,776 documents retrieved from the Scopus database regarding Molecular Diagnostics for Antibiotic Resistance Detection reveals a clear shift from limited early research interest to an intensive global academic focus. The initial period, spanning from 1991 to the late 1990s, was characterized by minimal publication volume (starting with only 2 documents in 1991 and remaining below 70 documents per year), indicating that the topic was in an embryonic phase where molecular technologies were still in early development within medical laboratories. However, entering the 2000s, the research began to show steady growth, surpassing 100 documents in 2003 (130 documents) and continuing to rise to 533 documents by 2015. The most significant exponential growth period occurred over the last decade (2015–2024), where the number of publications surged drastically from 533 to over 1,100 documents per year. The peak was recorded in 2024 with **1,105** publications, confirming that molecular diagnostics has become an essential research area and a top priority in addressing the global health crisis related to antimicrobial resistance (AMR).

The study by David et al. pioneered this field, specifically discussing the molecular analysis of retroviral transduction in chronic myelogenous leukemia [6]. The literature on molecular diagnostics for antibiotic resistance detection has evolved from fundamental studies of molecular mechanisms, such as enzymatic degradation and gene characterization [7][8][9], toward a strong emphasis on rapid diagnostic approaches like PCR, microarrays, and Next-Generation Sequencing [10][11][12]. Recent research frontiers highlight the integration of emerging technologies, including CRISPR-based systems [13], isothermal amplification [13], and microfluidic biosensors [14], alongside AI-enhanced data interpretation to support antimicrobial stewardship. Current trends reflect a strategic shift toward decentralized Point-of-Care (POC) testing and the combination of genotypic and phenotypic data to guide therapy [15][16][17], though significant barriers to implementation such as high costs, technical complexity, and the need for clinical standardization remain critical challenges for global scalability [18][19].



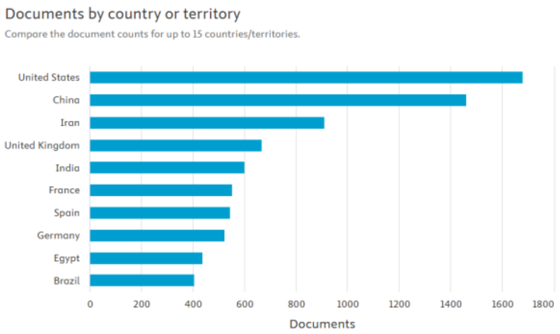
Source: Scopus database

Figure 2. Number of Molecular Diagnostics For Antibiotic Resistance Publications

The reviewed literature suggests that while research in this field began as early as the 1990s, the explosion of its urgency coincided with advancements in technologies such as Next-Generation Sequencing (NGS) and Point-of-Care Testing (POCT). The surge in publication output since 2019 (661 documents) to over 1,100 documents in 2024 highlights the scientific community's rapid response to the need for faster and more accurate detection methods. This phenomenon offers a crucial opportunity for academics and medical laboratory practitioners to delve into this vital topic with greater depth, particularly in the integration of Artificial Intelligence (AI) for interpreting bacterial genetic data. Studies focusing on this field are not merely academic exercises; they are essential for deepening our collective understanding of how various factors including the identification of new resistance genes, laboratory automation, and clinical validation of molecular methods influence long-term infection control effectiveness. Therefore, the intensification of research in this area is vital to support adaptive and strategic clinical decision-making in the era of precision medicine.

RQ2: Who are the key authors, institutions, and countries contributing to the most significant research in this field, and what are the patterns of their global research collaboration networks?

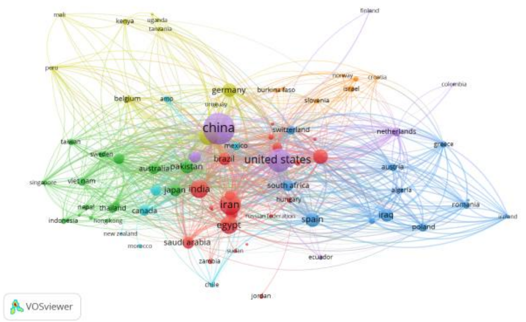
The global landscape of research on molecular diagnostics for antibiotic resistance is characterized by a strong concentration of scientific output in the United States and China, which lead with 1,678 and 1,459 documents respectively, highlighting their roles as primary hubs for biotechnological innovation. The significant contributions from Iran (908) and India (599), alongside European leaders such as the United Kingdom (665), France (549), and Spain (542), reflect a diverse international effort to address antimicrobial resistance across different healthcare systems. Furthermore, the inclusion of Egypt (436) and Brazil (402) among the top ten most productive countries underscores the global urgency of this field, demonstrating that the development of rapid molecular detection methods is a critical priority for both developed and emerging economies.



Source: Scopus database

Figure 3. Number of Articles by Country or Territory (Top 10 Countries)

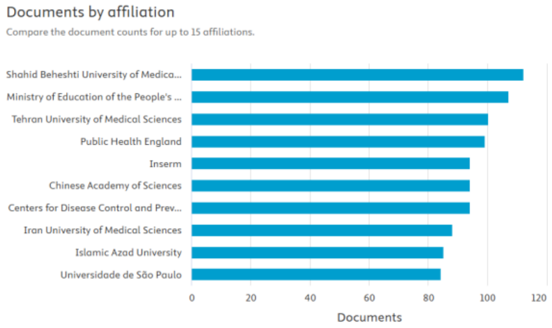
The VOSviewer visualization map illustrates a robust and interconnected global research network in the field of molecular diagnostics for antibiotic resistance, characterized by distinct geographical and thematic clusters. The United States and China emerge as the most prominent nodes, serving as central global hubs with the highest density of publications and extensive collaborative links across multiple continents. Regional clusters are clearly visible, such as the strong Middle Eastern network led by Iran, Saudi Arabia, and Egypt, as well as a significant European cluster featuring the United Kingdom, Spain, and the Netherlands. The intricate web of lines connecting these nodes signifies a high degree of international cross-border collaboration, indicating that the fight against antimicrobial resistance is a collective global effort. Furthermore, the presence of emerging research contributors like India, Brazil, and Australia within the core network reflects the widespread adoption of molecular technologies and a unified scientific response to this escalating public health crisis.



Source: VOSviewer software output

Figure 4. Visualization of countries in the network

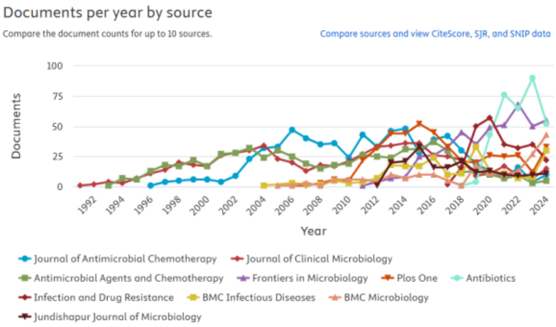
The distribution of research output by institutional affiliation reveals a highly competitive landscape dominated by prominent medical and governmental organizations, with Shahid Beheshti University of Medical Sciences leading the global contribution with 112 documents. The strong presence of Iranian institutions, including Tehran University of Medical Sciences (100) and Iran University of Medical Sciences (88), alongside major Chinese bodies such as the Ministry of Education (107) and the Chinese Academy of Sciences (94), highlights a concentrated regional expertise in molecular diagnostic research. Furthermore, the significant involvement of world-renowned public health agencies like Public Health England (99), Inserm (94), and the Centers for Disease Control and Prevention (94) underscores the strategic importance of this field in national security and global health surveillance. Collectively, these top ten affiliations, including the Universidade de São Paulo (84), represent the core institutional pillars driving the advancement of rapid detection technologies for antibiotic resistance across diverse clinical and academic settings.



Source: Scopus database

Figure 5. Number of Articles by Affiliation (Top 10 Affiliation)

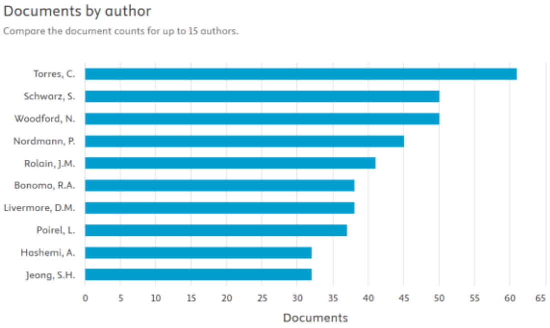
The distribution of publication sources reveals a highly specialized academic landscape, with the Journal of Antimicrobial Chemotherapy and the Journal of Clinical Microbiology emerging as the primary authorities, contributing 683 and 634 documents respectively. The prominence of high-impact journals such as Antimicrobial Agents and Chemotherapy (601) and Frontiers in Microbiology (453) underscores the rigorous clinical and microbiological standards required for research in molecular diagnostics. Additionally, the significant presence of open-access and multidisciplinary platforms like PLoS One (451) and Antibiotics (331), alongside specialized titles such as Infection and Drug Resistance (249) and the Jundishapur Journal of Microbiology (193), reflects the global and accessible nature of this field. Collectively, these top ten sources represent the core intellectual outlets for advancing the detection and management of antibiotic resistance within the medical laboratory community.



Source: Scopus database

Figure 6. Number of Articles by Sources (Top 10 Sources)

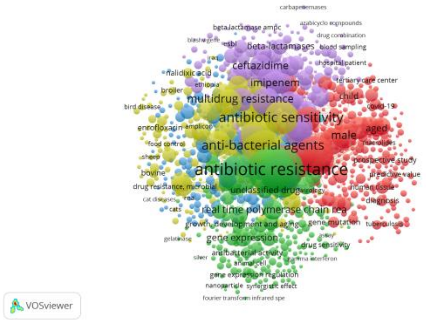
The concentration of scholarly expertise in the field of molecular diagnostics for antibiotic resistance is spearheaded by a group of highly prolific researchers, with Torres, C. leading the global contribution with 61 documents. The significant impact of authors such as Schwarz, S. (50), Woodford, N. (50), and Nordmann, P. (45) reflects a sustained commitment to advancing the understanding of resistance mechanisms and diagnostic innovation. Furthermore, the inclusion of influential experts like Rolain, J.M. (41), Bonomo, R.A. (38), and Livermore, D.M. (38), alongside Poirel, L. (37), Hashemi, A. (32), and Jeong, S.H. (32), highlights a geographically diverse intellectual leadership. Collectively, these top ten authors represent the primary scientific drivers whose extensive publications provide the foundational frameworks for modern clinical microbiology and global antimicrobial resistance surveillance.



Source: Scopus database

Figure 7. Number of Articles by Authors (Top 10 Authors)

The analysis of author keywords provides a profound overview of the intellectual structure and primary focus within the molecular diagnostics literature. The keyword co-occurrence analysis reveals that Antibiotic Resistance holds the top position with the highest Total Link Strength (TLS) of 35,836, confirming its role as the central theme and primary bridge connecting all research clusters within this dataset. The significant link strength of the keywords Genetics (23,515) and Polymerase Chain Reaction (21,067) indicates that the literature's core focus lies in identifying the genetic mechanisms of resistance through nucleic acid amplification methods. Furthermore, the emergence of terms such as Antibiotic Sensitivity (22,977) and Anti-bacterial Agents (20,696) reflects a robust correlation between molecular diagnosis and phenotypic susceptibility testing in clinical laboratory practice. Overall, the high TLS in the Human (25,536) category compared to the overlapping Nonhuman (26,643) data in the VOSviewer visualization suggests that this research encompasses a broad multidisciplinary scope, ranging from clinical patient applications to environmental surveillance and zoonosis within a One Health framework.



Source: VOSviewer software output

Figure 8. Co-occurrence framework and representation of key terms.

Table 1. Keywords by authors

Rank	Keywords	Total Link Strength
1	Antibiotic Resistance	35836
2	Article	35566
3	Nonhuman	26643
4	Human	25536
5	Genetics	23515
6	Antibiotic Sensitivity	22977
7	Microbiology	21101
8	Polymerase Chain Reaction	21067
9	Antiinfective Agent	21058
10	Anti-bacterial Agents	20696

Source: VOSviewer software output

RQ3: What are the specific molecular markers, diagnostic methodologies, and key research findings highlighted in the most influential literature regarding antibiotic resistance detection?

Advances in the identification of molecular markers have revolutionized antibiotic resistance detection, with a primary focus on resistance genes such as blaNDM, mcr-1, cfr, and various \beta -lactamase genes (TEM, SHV, OXA, CTX-M) associated with carbapenem and colistin resistance [20]. Beyond specific genes, mutations in target genes such as rdxA and pbp1A in Helicobacter pylori, alongside the role of mobile genetic elements in horizontal gene transfer, have become crucial focal points for understanding the dissemination of resistance in both clinical settings and agricultural ecosystems [21][22][23]. To detect these markers, diagnostic methodologies have evolved beyond conventional phenotypic and culture-based techniques, which tend to be slow [24][25]. Modern molecular techniques such as Next-Generation Sequencing (NGS) and multiplex PCR now enable the comprehensive and rapid mapping of multidrug resistance profiles, while innovative platforms based on CRISPR and microfluidics offer highly sensitive detection solutions with minimal sample requirements [26][20][27].

Recent research findings emphasize the integration of cutting-edge technologies to achieve rapid and decentralized diagnostics, where innovations such as lateral flow assays and electrochemical biosensors can reduce detection times to under 18 hours, making them ideal for point-of-care applications in resource-limited regions. The utilization of spectrometry, such as MALDI-TOF MS combined with Artificial Intelligence (AI) and machine learning applied to FTIR data, has also shown significant results in predicting resistance patterns dynamically and accurately [28]. Beyond clinical aspects, the literature highlights the importance of environmental surveillance in tracking resistance reservoirs within aquatic ecosystems and livestock, particularly for high-priority pathogens such as Pseudomonas aeruginosa and Escherichia coli [29][30][23]. Collectively, these findings suggest that the integration of advanced genotypic methods and adaptive public health frameworks is essential for the effective management of antibiotic resistance in the future [16][31].

Table 2. Key markers, diagnostics, and findings in influential antibiotic resistance literature

No	Key markers, diagnostics, and findings in influential antibiotic resistance literature	Reference
1	The increasing prevalence of multi-drug	[32]

No	Key markers, diagnostics, and findings in influential antibiotic resistance literature	Reference
	resistant organisms in intra-abdominal infections necessitates effective diagnostic strategies, such as PCR-based systems, which provide rapid identification and 100% diagnostic accuracy to enable timely, targeted antibiotic therapy.	
2	The identification of E.coli strains harboring co-located resistance genes such as CTX-M-15 and TEM-1B alongside specific gyrA and parC mutations emphasizes the critical role of advanced molecular diagnostics in rapidly detecting complex resistance mechanisms to ensure timely and effective clinical management.	[33]
3	The prevalence of multidrug-resistant (MDR) species such as *Staphylococcus aureus* and *Klebsiella pneumoniae* accounting for 62.7% of hospital-derived isolates highlights a critical healthcare concern, where the integration of 16S rRNA sequencing and high-throughput qPCR reveals that mobile genetic elements (MGEs) significantly facilitate the abundant transfer of resistance genes across clinical departments.	[34]
4	The persistent resistance of S.uberis isolates most notably against Ceftriaxone (79.31%) and Tetracycline, which saw a linear increase to 20.4% over a decade underscores the critical role of biofilm formation in therapeutic failure and the potential for antibiofilm agents to enhance antibiotic efficacy by up to two times in vitro.	[35]
5	The detection of five key antibiotic resistance (AMR) gene targets *aph*, *varG*, *floR*, *qnrVC5*, and *almG* which confer resistance to critical antibiotic classes such as aminoglycosides, carbapenems, and polymyxins, underscores the clinical necessity for rapid diagnostic tools to prevent ineffective prescriptions for *Vibrio cholerae* strains. Furthermore, the high diagnostic accuracy of LAMP assays, with 34 out of 43 demonstrating an AMR class specificity greater than 97%, highlights their significant potential as a reliable and accessible methodology for real-time resistance profiling in infectious disease	[36]

No	Key markers, diagnostics, and findings in influential antibiotic resistance literature	Reference
	management.	

CONCLUSION

This bibliometric and SLR study underscores the dynamic and interdisciplinary nature of research in molecular diagnostics for antibiotic resistance detection. The findings demonstrate that while traditional molecular techniques like PCR remain foundational, the field is rapidly advancing toward high-throughput and portable technologies. The emergence of CRISPR, microfluidic biosensors, and AI-driven data interpretation represents the current research frontier, promising faster and more accurate diagnostic workflows.

However, a significant gap remains between technological innovation and routine clinical implementation. Challenges such as high operational costs, technical complexity, and the need for standardized clinical validation continue to hinder widespread adoption, particularly in resource-limited settings. The study also highlights the importance of a *One Health* approach, as resistance genes are increasingly tracked across human, animal, and environmental reservoirs.

The future of antibiotic resistance management lies in the successful integration of rapid molecular diagnostics with global antimicrobial stewardship programs. Collaborative international research and the development of cost-effective, decentralized testing platforms are essential to mitigate the global burden of AMR and ensure the longevity of the existing antibiotic arsenal.

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