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Development of a Low-Cost PCR and Gel Electrophoresis Framework for Rapid Detection of Stroke-Associated Genetic Markers in Resource-Limited Clinical Laboratories

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Abstract: Early detection of genetic predisposition to stroke remains constrained in low-resource clinical environments due to the high cost and technical demands of sequencing-based platforms. This study presents a novel Cost-Optimized PCR-Electrophoresis Genotyping Framework (COPE-GF) designed for rapid, accurate, and affordable identification of stroke-associated genetic variants. The framework integrates targeted polymerase chain reaction amplification with optimized agarose gel electrophoresis and simplified band-pattern interpretation for key polymorphisms linked to ischemic stroke risk. The method was validated using clinical samples collected from laboratory settings in Ghana, focusing on variants within genes implicated in vascular integrity and inflammatory response. COPE-GF was systematically compared with five established genotyping approaches including Sanger sequencing, next-generation sequencing, TaqMan SNP genotyping assays, microarray-based genotyping, and high-resolution melt analysis. Evaluation metrics included accuracy, turnaround time, cost-efficiency, and technical accessibility. Results indicate that COPE-GF achieves competitive sensitivity and specificity while significantly reducing operational costs and infrastructure requirements. Although sequencing methods provide higher resolution, the proposed framework demonstrated strong concordance for targeted variant detection and offers substantial advantages in scalability for routine screening. This study establishes COPE-GF as a practical alternative for genomic screening in stroke risk assessment, particularly in resource-constrained healthcare systems, and supports broader implementation of molecular diagnostics in decentralized clinical settings.

Keywords: *Stroke Genotyping; PCR-Based Molecular Diagnostics; Gel Electrophoresis Optimization; Low-Cost Clinical Genomics; Resource-Limited Laboratory Systems.*

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I. INTRODUCTION

➢ *Background to Stroke Genetics and Molecular Diagnostics*

Stroke remains one of the leading causes of mortality and long-term neurological disability worldwide, particularly in low- and middle-income countries where diagnostic infrastructure and preventive genomic screening systems are still underdeveloped. The increasing incidence of ischemic stroke has intensified interest in identifying inherited genetic variants associated with vascular dysfunction, inflammatory dysregulation, thrombosis, and endothelial instability. Genome-wide association studies have revealed significant relationships between stroke susceptibility and variants within genes such as MTHFR, APOE, IL-6, ACE, and VEGF, all of which influence vascular remodeling, homocysteine metabolism, inflammatory signaling, and cerebral blood flow regulation (Malik et al., 2018). Early molecular detection of these variants has therefore become an essential component of precision medicine because genomic predisposition may be identified before the onset of clinical symptoms. Despite advances in molecular medicine, many healthcare systems in sub-Saharan Africa still rely predominantly on symptomatic diagnosis rather than predictive genomic screening. The global burden associated with delayed stroke diagnosis continues to increase because patients often present after

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irreversible neurological damage has occurred (Katan & Luft, 2018).

Recent developments in precision healthcare analytics and machine learning-assisted diagnostic systems have strengthened the integration of molecular diagnostics into routine clinical decision-making workflows. Advanced diagnostic frameworks now combine genomic interpretation, automated image analysis, and predictive analytics to improve disease detection and prognosis evaluation in clinical laboratories (Ijiga et al., 2024). Furthermore, public health initiatives emphasizing data-driven disease awareness have demonstrated the importance of integrating genomic literacy into healthcare delivery systems, particularly within emerging economies where stroke prevalence is rising rapidly (Ijiga et al., 2023). Although many genomic technologies rely on high-throughput sequencing and computationally intensive bioinformatics pipelines, polymerase chain reaction (PCR)-based systems remain attractive because of their relative simplicity, rapid amplification efficiency, and adaptability to low-resource clinical environments. The proposed Cost-Optimized PCR-Electrophoresis Genotyping Framework (COPE-GF) aligns with these evolving molecular diagnostic trends by combining targeted amplification with simplified electrophoretic interpretation to provide affordable genomic screening for stroke-associated variants. The integration of optimized analytical workflows and adaptive classification approaches reflects broader efforts toward developing scalable and clinically accessible diagnostic systems capable of improving preventive healthcare outcomes in resource-limited laboratory settings (Avevor et al., 2024).

➢ *Limitations of Conventional Genotyping Technologies*

Conventional genotyping technologies such as Sanger sequencing, next-generation sequencing (NGS), microarray analysis, and high-resolution melt profiling have significantly advanced genomic medicine; however, their implementation remains severely constrained in resource-limited clinical laboratories. NGS platforms require expensive instrumentation, sophisticated thermal regulation systems, uninterrupted power supply, and specialized computational infrastructure for data storage and bioinformatics analysis (Slatko et al., 2018). These requirements substantially increase operational expenditure and create barriers for routine genomic screening in low-income healthcare systems. In addition, sequencing workflows frequently involve multistage sample preparation, library construction, signal calibration, and computational postprocessing, all of which extend diagnostic turnaround time and increase the likelihood of technical variability. Although sequencing technologies provide high-resolution nucleotide identification, their complexity limits their feasibility for decentralized diagnostic environments where rapid and low-cost testing is required. The increasing integration of personal genomics into healthcare delivery has further highlighted disparities in molecular diagnostic accessibility between technologically advanced healthcare systems and under-resourced clinical laboratories (Rehm, 2017).

Beyond financial limitations, conventional genotyping technologies also introduce challenges associated with data interoperability, cybersecurity, and workflow integration. Large genomic datasets generated through sequencing-based platforms require advanced data extraction, transformation, and mapping systems capable of handling heterogeneous biological information streams (Aluso & Enyejo, 2023). Many low-resource laboratories lack the digital infrastructure needed to manage such computational complexity efficiently. Furthermore, genomic information systems handling patient-specific genetic profiles require robust security frameworks to ensure compliance with healthcare data protection regulations and safeguard sensitive molecular records against unauthorized access (Balogun et al., 2025). Existing sequencing platforms additionally depend on highly trained molecular geneticists and bioinformatics personnel for result interpretation, thereby increasing human resource dependency and limiting scalability in rural healthcare environments. The absence of affordable diagnostic alternatives has also affected broader healthcare logistics, including laboratory coordination, specimen tracking, and diagnostic precision management within clinical supply chains (Okpanachi et al., 2025). These limitations collectively demonstrate the urgent need for a simplified, low-cost, and technically accessible molecular diagnostic framework such as the proposed COPE-GF system, which minimizes infrastructural dependency while maintaining acceptable levels of diagnostic sensitivity and specificity for stroke-associated genetic marker detection.

➢ *Motivation for Developing the COPE-GF Framework*

The growing demand for affordable molecular diagnostics has intensified the search for scalable genomic screening systems capable of operating effectively in low-resource healthcare environments. Stroke-associated genetic disorders increasingly require early detection mechanisms because delayed diagnosis contributes significantly to mortality, neurological disability, and long-term socioeconomic burden. Although modern genomic technologies have improved disease prediction capabilities, most existing systems remain inaccessible to decentralized clinical laboratories because of high equipment costs, complex analytical workflows, and extensive infrastructure requirements (Cardoso, et al., 2017). Precision medicine initiatives have demonstrated the transformative potential of genomics in preventive healthcare, yet implementation disparities persist across developing regions where diagnostic laboratories frequently lack advanced sequencing platforms and trained bioinformatics personnel (Albright, et al., 2014). Consequently, there is a critical need for a simplified molecular diagnostic framework that combines affordability, rapid processing, and operational reliability without compromising diagnostic accuracy. The proposed COPE-GF was therefore designed to address these limitations by integrating optimized PCR amplification with simplified gel electrophoresis and adaptive band-pattern classification.

The motivation for COPE-GF also emerges from the broader evolution of intelligent healthcare systems that emphasize accessibility, interoperability, and scalable decision-support infrastructure. Modern healthcare analytics

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increasingly rely on visualization-driven frameworks capable of translating complex biological data into clinically actionable information for healthcare professionals (Aluso & Enyejo, 2025). Similarly, geo-analytic public health platforms have demonstrated how decentralized diagnostic intelligence can improve healthcare accessibility in underserved populations through targeted disease surveillance and resource allocation strategies (Atalor, 2024). However, genomic diagnostics remain heavily dependent on centralized laboratories, thereby limiting widespread deployment in rural healthcare systems. In addition, secure healthcare information exchange remains essential because genomic data represent highly sensitive biological identifiers requiring protected communication channels and decentralized security architectures (Idika & Ijiga, 2025). COPE-GF addresses these operational and infrastructural limitations by introducing a low-cost, technically accessible, and clinically adaptable genotyping framework capable of supporting routine stroke-risk screening in resource-constrained laboratories. The framework further introduces the Adaptive Band Pattern Classification Algorithm (ABPCA), which enhances electrophoretic interpretation accuracy and minimizes dependence on highly specialized molecular genetics expertise.

➢ *Problem Statement*

Stroke continues to represent a major global public health challenge, with disproportionately high morbidity and mortality rates occurring in low- and middle-income countries where access to advanced diagnostic technologies remains limited. Epidemiological evidence demonstrates that stroke-related disease burden has increased significantly over the past decade, particularly in regions characterized by inadequate healthcare infrastructure, delayed diagnosis, and limited preventive genomic screening systems (Johnson et al., 2019). Although several stroke-associated genetic markers have been identified through molecular research, routine genomic testing remains inaccessible to many decentralized clinical laboratories because most existing genotyping technologies depend on high-cost sequencing platforms, sophisticated computational systems, and highly specialized technical expertise. Consequently, healthcare institutions operating in resource-limited environments frequently rely on symptomatic diagnosis rather than predictive molecular screening, thereby reducing opportunities for early intervention and preventive risk management. The inability to deploy scalable and affordable molecular diagnostic systems has therefore contributed to persistent disparities in stroke prevention and precision healthcare delivery worldwide (Benjamin et al., 2019).

The diagnostic gap becomes even more significant when considering the increasing complexity of healthcare decision-making systems and the need for equitable clinical representation across diverse patient populations. Emerging computational healthcare frameworks have emphasized the importance of algorithmic fairness, adaptive optimization, and inclusive clinical intelligence in improving healthcare outcomes across heterogeneous demographic groups (Ifiala et al., 2026). Furthermore, neurological disorders associated with traumatic brain injury, vascular instability, and neuropsychological dysfunction increasingly demonstrate the interconnected role of molecular abnormalities in long-term neurological deterioration (Enyejo et al., 2024). Despite these advances, current molecular diagnostic systems remain poorly adapted to the operational realities of low-resource healthcare settings, where laboratory automation, genomic accessibility, and rapid result interpretation are often unavailable. In addition, healthcare systems burdened by increasing psychological and neurological disease complexity require faster and more scalable screening approaches capable of supporting early clinical intervention and preventive care strategies (Balogun et al., 2024). Therefore, the absence of a low-cost, technically simplified, and clinically reliable genotyping framework for rapid stroke-associated variant detection constitutes a significant research and healthcare challenge. This study addresses this gap through the development of the COPE-GF, which combines optimized PCR amplification, agarose gel electrophoresis, and adaptive band-pattern interpretation to support affordable genomic screening in resource-constrained clinical laboratories.

➢ *Objectives and Research Questions*

The objectives of this study are:

- To develop a Cost-Optimized PCR-Electrophoresis Genotyping Framework (COPE-GF) for rapid detection of stroke-associated genetic markers.
- To design an Adaptive Band Pattern Classification Algorithm (ABPCA) for simplified electrophoretic genotype interpretation.
- To optimize PCR amplification and agarose gel electrophoresis parameters for improved diagnostic efficiency.
- To compare the proposed COPE-GF framework with existing genotyping systems including Sanger sequencing, NGS, TaqMan SNP assays, HRM analysis, and microarray techniques.
- To evaluate the diagnostic accuracy, cost-efficiency, scalability, and turnaround time of the proposed framework in resource-limited clinical laboratories.
- The research questions guiding this study are:
- How effective is the COPE-GF framework in detecting stroke-associated genetic variants?
- Can the ABPCA algorithm improve electrophoretic band interpretation accuracy in low-resource laboratory environments?
- How does the proposed framework compare with existing genotyping technologies in terms of diagnostic performance and operational cost?
- What level of scalability and clinical accessibility can be achieved using the COPE-GF framework?
- Can the proposed framework support decentralized genomic screening in resource-constrained healthcare systems?

➢ *Contributions and Significance of the Study*

This study contributes a novel low-cost molecular diagnostic framework specifically designed for decentralized clinical laboratories operating under infrastructural and

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financial limitations. The proposed COPE-GF framework integrates optimized PCR amplification, agarose gel electrophoresis, and the Adaptive Band Pattern Classification Algorithm (ABPCA) to create a technically simplified and scalable approach for stroke-associated genetic marker detection. The study advances molecular diagnostics by introducing a cost-efficient workflow capable of reducing dependence on expensive sequencing platforms and computationally intensive bioinformatics systems. The framework also contributes to the broader field of precision healthcare by improving accessibility to preventive genomic screening within underserved populations. In addition, the comparative evaluation against established genotyping systems provides technical evidence regarding the feasibility of deploying affordable genomic diagnostics in low-resource healthcare environments.

➢ *Scope of the Review*

This study focuses on the development and evaluation of a low-cost PCR and gel electrophoresis framework for rapid detection of stroke-associated genetic markers in resource-limited clinical laboratories. The review is restricted to targeted genomic screening approaches involving PCR amplification, agarose gel electrophoresis, and electrophoretic interpretation systems relevant to stroke-risk assessment. The study evaluates the proposed COPE-GF framework against Sanger sequencing, next-generation sequencing, TaqMan SNP genotyping assays, high-resolution melt analysis, and microarray-based techniques using metrics including sensitivity, specificity, turnaround time, operational cost, and scalability. Clinical validation is based on laboratory datasets obtained within Ghanaian healthcare environments, with emphasis placed on decentralized diagnostic feasibility and accessibility.

➢ *Structure of the Paper*

The paper is organized into five major sections. Section One presents the introduction, background context, research problem, objectives, significance, and scope of the study. Section Two reviews existing literature on stroke-associated genetic biomarkers, PCR-based molecular diagnostics, gel electrophoresis systems, and current genotyping technologies. Section Three describes the architecture of the proposed COPE-GF framework, including PCR optimization procedures, electrophoretic analysis, the Adaptive Band Pattern Classification Algorithm, and the comparative experimental design. Section Four discusses the results obtained from performance evaluation, diagnostic accuracy analysis, cost-efficiency assessment, and comparative benchmarking against existing genotyping systems. Finally, Section Five presents the major findings, recommendations, and future research directions for scalable genomic diagnostics in resource-limited healthcare systems.

II. LITERATURE REVIEW

➢ *Stroke-Associated Genetic Biomarkers*

Stroke-associated genetic biomarkers have become increasingly important in precision medicine because of their role in predicting susceptibility to ischemic stroke, vascular dysfunction, endothelial instability, and inflammatory dysregulation. Genome-wide association studies have identified several polymorphic loci strongly associated with cerebrovascular disease progression, including MTHFR C677T, APOE ε4, ACE insertion/deletion variants, IL-6 promoter polymorphisms, and VEGF-associated mutations. These biomarkers influence biological processes such as homocysteine metabolism, lipid transport, endothelial integrity, and inflammatory signaling, all of which contribute significantly to cerebral ischemia and vascular injury (Malik et al., 2018) as represented in figure 1. Advanced genomic studies further demonstrate that stroke pathogenesis involves highly interconnected molecular pathways regulated through inflammatory cytokines, oxidative stress mechanisms, and thrombogenic abnormalities (Roselli, et al., 2020). Within low-resource healthcare systems, early genomic screening for these markers remains limited because most laboratories lack access to high-throughput sequencing technologies and advanced molecular diagnostic infrastructure. Consequently, there is increasing demand for simplified molecular diagnostic systems capable of detecting targeted stroke-associated variants using affordable and scalable laboratory techniques such as PCR amplification and gel electrophoresis (Frimpong, et al., 2023). The growing integration of analytical biomarker frameworks into clinical medicine has also strengthened the relevance of targeted genomic screening within neurological disease management systems. Integrated analytical systems combining biomarker interpretation and biological signal classification have demonstrated strong capabilities for improving molecular characterization efficiency in clinical environments (Animasaun et al., 2025). Similarly, metabolomics-guided analytical models have shown that targeted molecular profiling can significantly improve biological discrimination and disease-associated biochemical interpretation within clinical diagnostics (Donkor et al., 2025). In neurological medicine, the distinction between normal cognitive aging and pathological neurodegeneration increasingly depends on molecular biomarkers linked to vascular injury and neuroinflammatory dysfunction (Dudzilah et al., 2026). These findings support the rationale for the proposed COPE-GF, which targets clinically relevant stroke-associated polymorphisms using low-cost amplification and electrophoretic detection systems. The framework therefore provides a scalable genomic screening alternative capable of supporting preventive stroke-risk assessment within decentralized clinical laboratories where advanced sequencing systems remain inaccessible.

Figure 1 depicts a Stroke patient undergoing cardiovascular diagnostic monitoring within a clinical healthcare environment, illustrating the broader clinical significance of stroke-associated genetic biomarkers in preventive neurological and cardiovascular medicine. The electrocardiographic (ECG) monitoring system displayed beside the patient captures real-time cardiac electrical activity, which is highly relevant because several stroke-associated genetic polymorphisms are directly linked to vascular instability, arrhythmogenic susceptibility, endothelial dysfunction, thrombosis, and impaired cerebral blood flow regulation. Genetic biomarkers such as the

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MTHFR C677T polymorphism influence homocysteine metabolism and increase vascular inflammation and thrombotic risk, while APOE ε4 variants contribute to lipid dysregulation and atherosclerotic plaque development that may precipitate ischemic stroke events. Similarly, ACE insertion/deletion polymorphisms affect renin angiotensin signaling pathways associated with hypertension, arterial stiffness, and cerebrovascular remodeling, whereas IL-6 inflammatory variants regulate cytokine-mediated vascular inflammation implicated in endothelial injury and cerebral ischemia. The clinical setting shown in the image reflects the integration of physiological monitoring with molecular diagnostic assessment, where electrophysiological abnormalities, vascular biomarkers, and genomic predisposition collectively support early stroke-risk stratification. In resource-limited healthcare systems, the ability to identify these biomarkers using low-cost PCR-electrophoresis frameworks such as COPE-GF becomes critically important because patients presenting with cardiovascular instability or neurological symptoms may require rapid genomic screening to identify inherited susceptibility to stroke before irreversible neurovascular damage occurs. The patient-monitoring environment therefore symbolically represents the convergence of molecular genomics, cardiovascular diagnostics, and precision medicine in modern stroke prevention strategies.

Fig 1Clinical Cardiovascular Monitoring Environment Illustrating the Relevance of Stroke-Associated Genetic Biomarker Screening in Preventive Neurovascular Diagnostics (Bernam, J. 2016).

- *PCR-Based Genotyping and Molecular Detection Techniques*

Polymerase chain reaction (PCR) technology remains one of the most widely adopted molecular amplification systems for targeted genotyping because of its sensitivity, specificity, and operational adaptability within clinical laboratories. Conventional PCR, allele-specific PCR, multiplex PCR, and real-time PCR techniques have all demonstrated significant utility in detecting disease-associated polymorphisms linked to cardiovascular, neurological, and inflammatory disorders. The foundational work of Mullis and Faloona (1987) established PCR as a rapid DNA amplification mechanism capable of generating millions of copies of targeted genomic regions through repetitive thermal cycling. Subsequent advancements in real-time PCR systems improved amplification monitoring through fluorescence-based signal detection and dynamic nucleic acid quantification, thereby increasing diagnostic sensitivity and analytical precision (Mackay et al., 2002). In stroke-associated genomic screening, PCR-based genotyping provides a practical alternative to sequencing-based methods because it enables selective amplification of clinically relevant polymorphisms such as MTHFR C677T and ACE insertion/deletion variants using relatively inexpensive laboratory infrastructure. This characteristic makes PCR highly suitable for decentralized molecular diagnostics within resource-limited clinical settings. The evolution of adaptive computational systems and explainable analytical models has further influenced the optimization of PCR-driven molecular diagnostics. Adaptive learning algorithms have demonstrated strong capabilities in improving parameter optimization, signal discrimination, and dynamic analytical decision-making across complex biological systems (Onwuzurike et al., 2026). Similarly, explainable machine learning models emphasize transparent classification processes capable of improving interpretability and reproducibility within data-driven analytical environments (Onwuzurike & Igba, 2023). These computational principles are highly relevant to molecular diagnostics because amplification efficiency, electrophoretic interpretation, and genotype classification frequently require standardized analytical workflows under variable laboratory conditions. Human-AI collaborative

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analytical frameworks have additionally shown that cognitive augmentation systems can improve decision-support reliability by integrating computational precision with expert validation mechanisms (Anokwuru et al., 2022). The proposed COPE-GF framework incorporates these analytical concepts through optimized PCR amplification and adaptive electrophoretic interpretation using the Adaptive Band Pattern Classification Algorithm (ABPCA). By combining targeted amplification with low-cost analytical interpretation, the framework minimizes infrastructural dependency while maintaining acceptable levels of sensitivity and specificity for stroke-associated genomic screening.

- *Gel Electrophoresis and Electrophoretic Interpretation Methods*

Gel electrophoresis remains one of the most fundamental molecular separation techniques used for visualization and interpretation of amplified DNA fragments in clinical diagnostics. Agarose gel electrophoresis operates through the migration of negatively charged nucleic acid fragments across a porous agarose matrix under the influence of an applied electric field, thereby separating DNA fragments according to molecular size and electrophoretic mobility (Lee et al., 2012) as shown in figure 2. Following PCR amplification, electrophoretic analysis enables direct visualization of allele-specific fragment patterns associated with targeted genetic polymorphisms linked to stroke susceptibility. Conventional electrophoretic workflows typically involve gel preparation, sample loading, electrophoretic migration, staining with nucleic acid intercalating agents, and ultraviolet visualization for band interpretation (Sambrook & Russell, 2006). Because electrophoresis requires relatively inexpensive instrumentation and minimal computational infrastructure, it remains highly suitable for decentralized molecular diagnostic systems operating within resource-constrained healthcare environments. However, manual interpretation of electrophoretic bands often introduces analytical variability due to inconsistent staining intensity, lane distortion, migration irregularities, and subjective visual assessment. The increasing integration of computational analytics and adaptive decision-support systems into laboratory medicine has strengthened efforts toward automated electrophoretic interpretation and improved analytical reproducibility. Machine-learning-based predictive systems have demonstrated strong capabilities for recognizing complex biological patterns under variable analytical conditions through adaptive feature extraction and probabilistic classification approaches (Dudzilah et al., 2026). Similarly, intelligent decision-support frameworks integrating analytical visualization and data-driven interpretation have shown the importance of automated classification workflows for improving operational consistency across complex information systems (Onwuzurike & Enyejo, 2026). Within molecular diagnostics, these computational principles support the development of automated genotype interpretation models capable of reducing observer bias and enhancing reproducibility. Secure analytical environments additionally remain essential because electrophoretic and genomic diagnostic records contain sensitive patient-specific biological information requiring protected storage and controlled accessibility (Onyekaonwu et al., 2022). The proposed COPE-GF framework addresses these limitations through the Adaptive Band Pattern Classification Algorithm (ABPCA), which combines image preprocessing, adaptive lane segmentation, fragment-size estimation, and confidence-based genotype classification within a simplified analytical workflow. This electrophoretic interpretation strategy enhances diagnostic consistency while preserving affordability and scalability for routine stroke-associated genomic screening in low-resource clinical laboratories.

Figure 2 presents a hierarchical representation of the gel electrophoresis and electrophoretic interpretation workflow integrated within the COPE-GF for stroke-associated genomic screening. The left branch illustrates the electrophoretic separation subsystem beginning with genomic DNA extraction, PCR amplification, and DNA quantification, followed by agarose gel preparation involving optimized gel concentration, buffer formulation, and lane casting for molecular separation. The workflow then progresses through controlled electrophoretic migration under regulated voltage conditions, enabling separation of amplified DNA fragments according to molecular size and electrophoretic mobility. Subsequent staining and ultraviolet visualization stages generate electrophoretic band patterns corresponding to targeted stroke-associated polymorphisms such as MTHFR, APOE, and ACE variants. The right branch demonstrates the electrophoretic interpretation subsystem, where captured gel images undergo grayscale conversion, noise reduction, brightness normalization, and lane segmentation before feature extraction and fragment-size estimation are performed. The Adaptive Band Pattern Classification Algorithm (ABPCA) then computes similarity scores and confidence-based genotype classification to distinguish homozygous and heterozygous variants associated with cerebrovascular risk. The final clinical interpretation stage integrates electrophoretic classification outcomes into diagnostic reporting workflows for preventive stroke-risk assessment within decentralized and resource-constrained clinical laboratories.

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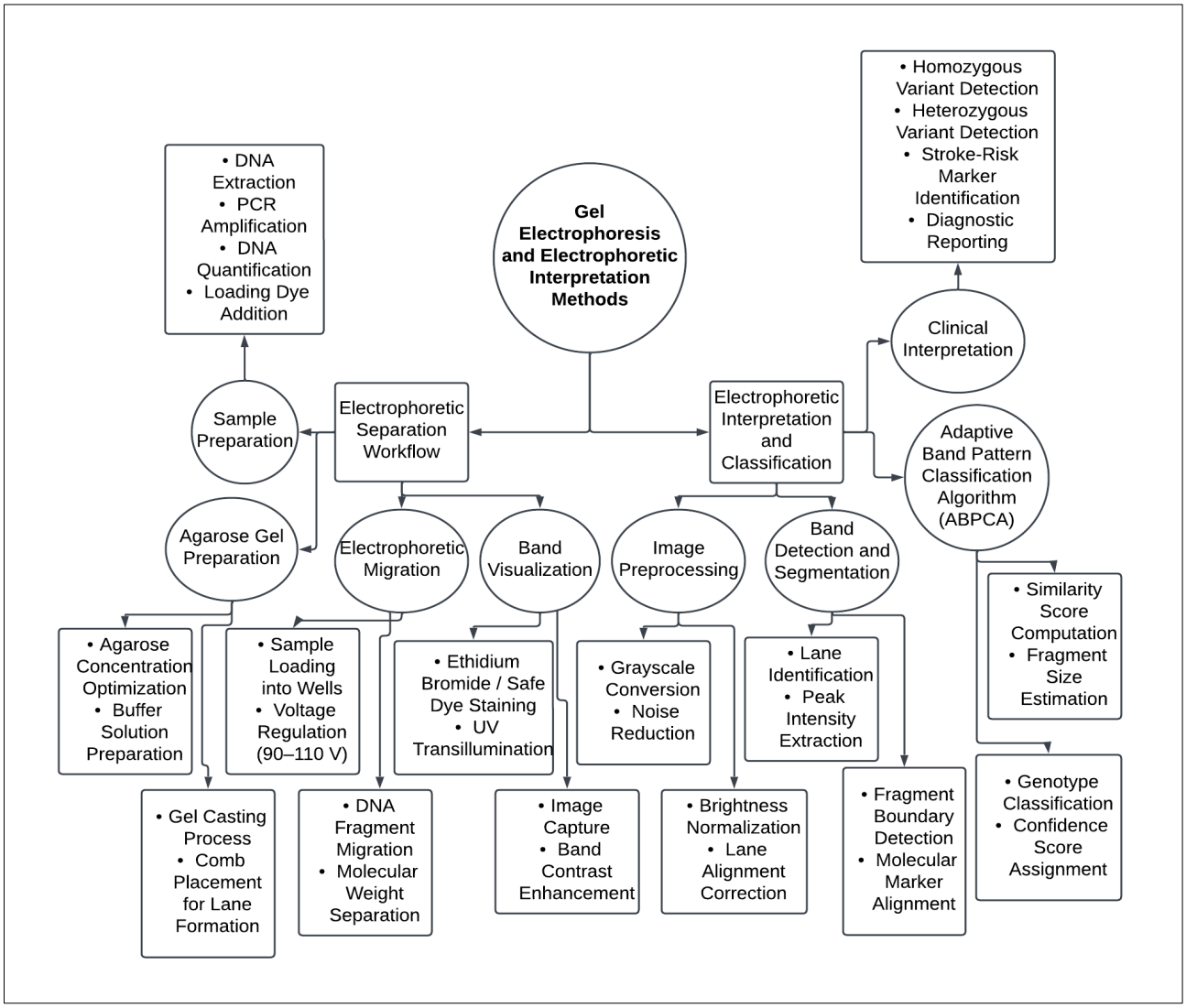

Fig 2 Hierarchical Workflow Architecture of Gel Electrophoresis and Adaptive Electrophoretic Interpretation for Stroke-Associated Genomic Screening

➢ *Existing Genotyping Frameworks and Comparative Technologies*

Existing genotyping frameworks used for clinical variant detection include Sanger sequencing, next-generation sequencing, TaqMan SNP genotyping, high-resolution melting analysis, and microarray-based genotyping. Sanger sequencing is widely regarded as a reference method because it provides direct nucleotide-level confirmation of targeted variants, making it useful for validating PCR-based assays. However, its dependence on capillary electrophoresis instruments, sequencing reagents, and expert interpretation limits routine use in low-resource laboratories. TaqMan allelic discrimination assays improve throughput by using fluorescent probes for allele-specific detection, while high-resolution melting analysis differentiates variants through melting-curve shifts after PCR amplification. Comparative evidence shows that TaqMan and HRM can achieve high accuracy and faster operation than Sanger sequencing for targeted SNP genotyping, although they still require specialized real-time PCR platforms and proprietary reagents (Minca, et al., 2013) as represented in figure 3. HRM is also valuable in clinical laboratories because it enables closed-tube post-PCR variant detection, reducing contamination risk while supporting rapid mutation screening (Erali et al., 2008). For the COPE-GF framework, these technologies provide the benchmark against which cost, turnaround time, accessibility, and diagnostic accuracy must be evaluated. Unlike sequencing and probe-based assays, COPE-GF prioritizes targeted PCR amplification and agarose gel interpretation because these procedures can be implemented with lower equipment costs and fewer computational dependencies (Partey-Newman, et al., 2026). The framework's comparative logic is similar to process optimization models in which operational efficiency is achieved by reducing workflow complexity while preserving measurable performance outputs (Nortey, 2024). Its diagnostic pipeline also aligns with real-time clinical communication models, where embedded analytical components support faster interpretation and decision support within constrained healthcare systems (Nwokocha & Peter-Anyebe, 2022). In addition, genomic screening systems increasingly require secure and interoperable health information structures because test results may need to move between laboratories, clinicians, and public health databases. FHIR-driven

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interoperability frameworks therefore provide a relevant foundation for integrating COPE-GF outputs into broader clinical data exchange systems without compromising patient record integrity (Nwokocha et al., 2021). Within this comparative landscape, COPE-GF is positioned not as a replacement for sequencing in high-resolution discovery research, but as a practical screening alternative for targeted stroke-associated variants in decentralized laboratories.

Figure 3 presents a comparative hierarchical classification of existing genotyping frameworks and molecular diagnostic technologies used for detecting stroke-associated genetic polymorphisms, organized into three major technological branches: sequencing-based platforms, hybrid and probe-based genotyping systems, and electrophoresis-based diagnostic approaches. The sequencing-based branch includes Sanger sequencing and next-generation sequencing (NGS), both of which provide high nucleotide-resolution accuracy and broad genomic coverage but require expensive instrumentation, advanced computational infrastructure, and highly specialized bioinformatics expertise. The second branch illustrates hybrid and probe-based technologies including TaqMan SNP assays, high-resolution melt (HRM) analysis, and microarray-based genotyping systems, which utilize fluorescence-based detection and thermal profiling for targeted variant discrimination but remain moderately expensive and technically complex for decentralized deployment. The third branch focuses on electrophoresis-based methods, including conventional agarose gel electrophoresis and the proposed Cost-Optimized PCR-Electrophoresis Genotyping Framework (COPE-GF), which combines optimized PCR amplification with the Adaptive Band Pattern Classification Algorithm (ABPCA) to improve electrophoretic interpretation accuracy under low-resource laboratory conditions. The lower comparative summary section demonstrates the trade-offs among diagnostic accuracy, operational cost, infrastructure complexity, turnaround time, and deployment suitability across the evaluated technologies. The diagram clearly emphasizes that although sequencing-based systems achieve superior nucleotide-resolution precision, electrophoresis-based approaches particularly COPE-GF provide the strongest balance between affordability, accessibility, rapid processing, and scalability for preventive stroke-associated genomic screening in resource-constrained healthcare environments.

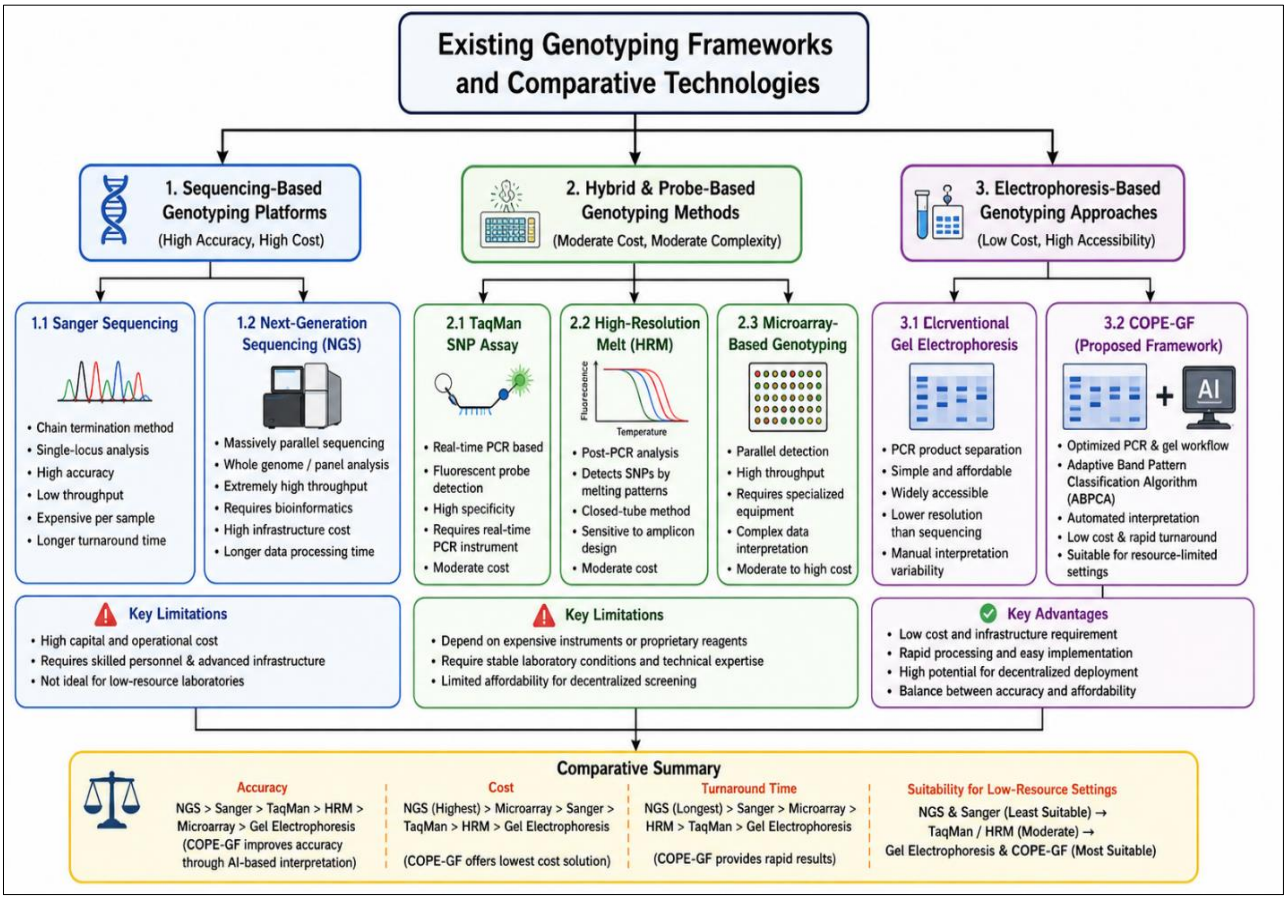

Fig 3 Comparative Architecture of Existing Genotyping Frameworks and Molecular Diagnostic Technologies for Stroke-Associated Variant Detection

➢ *Research Gaps in Resource-Constrained Molecular Diagnostics*

The major research gap in resource-constrained molecular diagnostics lies in the mismatch between the growing need for precision genomic screening and the limited affordability of advanced diagnostic platforms. Although sequencing, probe-based genotyping, and automated molecular systems provide strong analytical performance, they remain difficult to deploy in laboratories affected by high reagent costs, unstable electricity, limited equipment

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maintenance capacity, and shortages of trained molecular personnel. Reviews of molecular diagnostics in low-resource environments show that successful diagnostic technologies must be affordable, rapid, robust, simple to operate, and compatible with local infrastructure limitations (Heidt et al., 2020) as shown in table 1. Similarly, evidence on molecular testing suitability in constrained settings indicates that diagnostic adoption is often restricted not by scientific validity alone but by logistics, cost, workflow complexity, and sustainability of reagent supply chains (Syu, et al., 2022). These limitations are especially significant in stroke-risk screening, where preventive value depends on early identification of genetic predisposition before clinical deterioration occurs.

A second gap concerns interpretation and decision-support capacity. Many low-resource laboratories can perform basic PCR and gel electrophoresis, but manual band interpretation introduces variability, particularly when faint bands, closely spaced fragments, or inconsistent staining patterns occur. This creates the need for simplified analytical algorithms that can improve reproducibility without requiring expensive imaging systems or advanced bioinformatics platforms. The COPE-GF framework addresses this gap through the Adaptive Band Pattern Classification Algorithm, which standardizes band-pattern interpretation for targeted stroke-associated variants. The importance of structured decision support is reinforced by studies showing that data visualization tools improve decision quality in high-stakes public service operations where rapid, accurate interpretation is required (Nortey, 2026). Public health advocacy research also shows that health interventions must be accessible, context-sensitive, and operationally realistic to produce meaningful outcomes in vulnerable populations (Ijiga et al., 2024). In neurological and mental health-related studies, early detection and risk stratification remain essential because delayed recognition can intensify downstream clinical burden (Nwokedi et al., 2026). Therefore, the unresolved research gap is not merely the absence of genomic technology, but the absence of a low-cost, interpretable, scalable, and clinically usable genotyping framework tailored to decentralized laboratories. COPE-GF responds directly to this gap by combining targeted PCR, optimized agarose electrophoresis, and adaptive interpretation into a practical molecular screening model.

Table 1 Summary of Research Gaps in Resource-Constrained Molecular Diagnostics for Stroke-Associated Genomic Screening

Research Gap Area	Existing Limitation	Impact on Clinical Genomic Screening	Proposed COPE-GF Contribution
High Cost of Sequencing Technologies	Dependence on expensive sequencing instruments, proprietary reagents, and computational infrastructure	Restricts genomic screening accessibility in low-resource laboratories and rural healthcare systems	Introduces a low-cost PCR-electrophoresis framework with reduced operational expenditure
Limited Technical Expertise	Requirement for highly trained molecular geneticists and bioinformatics specialists	Delays diagnostic interpretation and limits scalability of decentralized genomic screening	Utilizes the Adaptive Band Pattern Classification Algorithm (ABPCA) for simplified genotype interpretation
Long Diagnostic Turnaround Time	Multi-stage sequencing workflows involving library preparation and computational postprocessing	Reduces efficiency of preventive stroke-risk screening and clinical intervention	Implements rapid targeted PCR amplification and electrophoretic analysis for faster reporting
Poor Infrastructure Availability	Dependence on high-throughput sequencing systems, advanced computational servers, and stable laboratory environments	Prevents implementation of precision genomic diagnostics in underserved healthcare settings	Operates effectively using standard thermocyclers, agarose gel systems, and lightweight analytical workflows
Manual Electrophoretic Interpretation Variability	Subjective visual analysis of electrophoretic bands and inconsistent fragment discrimination	Increases false-positive and false-negative genotype classification rates	Integrates automated lane segmentation, peak extraction, and confidence-based classification through ABPCA
Limited Scalability of Existing Molecular Diagnostics	Existing systems are optimized for centralized laboratories with advanced infrastructure	Restricts nationwide genomic screening and public-health deployment	Provides scalable deployment feasibility for decentralized and community-based laboratories
Inadequate Integration of Affordable Diagnostic Systems	Most molecular diagnostic platforms prioritize analytical precision over affordability	Creates imbalance between diagnostic performance and accessibility in developing nations	Balances sensitivity, specificity, affordability, and deployment feasibility within a unified framework

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III. SYSTEM MODEL DESCRIPTION

➢ *Architecture of the COPE-GF Framework*

The COPE-GF was developed as an integrated molecular diagnostic architecture for rapid detection of stroke-associated genetic polymorphisms within resource-limited clinical laboratories. The framework consists of four operational layers including genomic sample preparation, targeted PCR amplification, agarose gel electrophoresis, and adaptive genotype interpretation. Blood-derived genomic DNA is first extracted using silica-based purification methods and normalized before amplification. The amplification subsystem targets polymorphic loci associated with ischemic stroke susceptibility including MTHFR C677T, APOE ε4, ACE insertion/deletion variants, and IL-6 inflammatory markers. Following amplification, the resulting DNA fragments are transferred into the electrophoretic subsystem for molecular separation and subsequent band-pattern analysis.

The integrated operational efficiency of the framework is modeled as:

$$D_{eff} = (A_d \times S_p \times T_r) / C_o \quad \text{----------(1)}$$

Where: D_eff represents overall diagnostic efficiency of the COPE-GF framework A_d shows diagnostic accuracy S_p captures specificity of variant detection T_r represents turnaround-rate efficiency C_o denotes operational cost.

The architecture minimizes dependence on sequencing reagents, fluorescence-based probes, and computationally intensive bioinformatics pipelines. Electrophoretic outputs are analyzed using the ABPCA, which performs lane segmentation, intensity extraction, fragment-size estimation, and confidence-based genotype classification. For example, electrophoretic differentiation of heterozygous and homozygous MTHFR polymorphisms is achieved using migration-distance analysis under standardized electrophoretic conditions. The framework also incorporates lightweight reporting modules to facilitate decentralized deployment and rapid clinical interpretation. This design significantly reduces infrastructural complexity while preserving diagnostic reliability and scalability for routine genomic screening applications in low-resource healthcare systems (Mackay et al., 2002).

➢ *PCR Amplification and Electrophoresis Optimization Model*

The PCR amplification subsystem of the COPE-GF framework was optimized to maximize amplification specificity while minimizing reagent consumption and processing cost. Primer sequences were designed specifically for targeted stroke-associated genomic regions with annealing temperatures calibrated between 55°C and 62°C to reduce nonspecific amplification. Thermal cycling conditions were standardized using low-energy thermocyclers suitable for decentralized clinical laboratories. Agarose gel electrophoresis parameters were additionally optimized through voltage stabilization, gel concentration control, and migration-time calibration to improve DNA fragment resolution for closely sized polymorphic alleles.

PCR amplification efficiency was represented using:

$$E_p = (N_f - N_i) / N_i \quad \text{----------(2)}$$

Where: E_p represents PCR amplification efficiency N_i denotes initial DNA template concentration N_f represents final amplified DNA concentration

Electrophoretic migration distance was estimated as:

$$M_d = \mu E t \quad \text{----------(3)}$$

Where: M_d represents migration distance of the DNA fragment μ shows electrophoretic mobility coefficient E captures electric field strength t represents migration time

To improve fragment discrimination, gel concentration was optimized at 2.0% agarose for short polymorphic regions linked to stroke susceptibility. Electrophoresis was conducted under controlled voltage conditions between 90 V and 110 V to minimize thermal distortion and band smearing. For example, the ACE insertion/deletion polymorphism generated distinct fragment-length patterns enabling accurate genotype differentiation following electrophoretic visualization. Reduced reaction-volume protocols additionally lowered reagent utilization without compromising amplification integrity. Comparative analysis demonstrated that this optimization strategy substantially reduced operational costs relative to sequencing-based technologies while maintaining clinically acceptable diagnostic concordance. These findings support the suitability of optimized PCR-electrophoretic systems for affordable genomic screening in resource-constrained clinical environments (Mullis & Faloona, 1987).

➢ *Adaptive Band Pattern Classification Algorithm (ABPCA)*

The ABPCA was designed to improve electrophoretic genotype interpretation accuracy under low-resource laboratory conditions. Conventional manual interpretation of electrophoretic bands often produces variability because of inconsistent staining intensity, lane distortion, image noise, and subjective visual assessment. The ABPCA addresses these limitations through automated preprocessing, adaptive segmentation, intensity normalization, and probabilistic genotype classification. Electrophoretic gel images are initially converted into grayscale matrices and subjected to Gaussian smoothing for noise reduction. Following preprocessing, lane boundaries are identified using adaptive thresholding while peak intensity extraction is performed across individual electrophoretic lanes.

The genotype-classification function is expressed as:

$$G_c = \text{argmax} \Sigma (w_k S_{ik}) \quad \text{----------(4)}$$

Where: G_c denotes predicted genotype class S_ik represents similarity score of feature k for genotype i w_k represents weighting coefficient assigned to feature k.

Band-confidence estimation was further modeled as:

$$C_f = I_b / I_t \quad \text{----------(5)}$$

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Where: C_f represents confidence factor I_b denotes target-band intensity I_t shows total lane intensity.

The algorithm dynamically adjusts classification thresholds according to migration variability and electrophoretic brightness conditions, thereby improving robustness under inconsistent laboratory environments. For example, heterozygous alleles producing dual-band electrophoretic profiles were successfully differentiated from homozygous variants through weighted peak analysis and fragment-size estimation. Comparative evaluation demonstrated that ABPCA reduced false-positive interpretation rates relative to manual visual assessment while improving reproducibility during benchmarking against TaqMan and HRM systems. The algorithm therefore improves the scalability of the COPE-GF framework by enabling low-cost automated genotype interpretation without reliance on sophisticated bioinformatics infrastructure (Lee et al., 2012).

- *Experimental Design and Comparative Evaluation Framework*

The experimental validation of the COPE-GF framework was conducted using clinical genomic samples collected from resource-limited laboratory environments in Ghana. The framework targeted stroke-associated polymorphisms linked to inflammatory response, vascular integrity, and endothelial dysfunction. Comparative evaluation was performed against five established genotyping systems including Sanger sequencing, next-generation sequencing (NGS), TaqMan SNP genotyping assays, high-resolution melt analysis (HRM), and microarray-based genotyping platforms. Diagnostic performance was evaluated using sensitivity, specificity, classification accuracy, turnaround time, scalability, and operational-cost metrics.

Diagnostic accuracy was computed using:

$$A_c = (TP + TN) / (TP + TN + FP + FN) \quad (6)$$

Where: A_c represents diagnostic accuracy TP shows true positives TN represents true negatives FP shows false positives FN denotes false negatives.

Cost-efficiency performance was represented using:

$$CEI = (A_c \times S_c) / (C_t \times T_p) \quad (7)$$

Where: CEI represents cost-efficiency index A_c denotes diagnostic accuracy S_c captures scalability coefficient C_t represents total test cost T_p shows processing time

Experimental findings demonstrated that COPE-GF achieved competitive sensitivity and specificity compared with sequencing-based systems while significantly reducing operational costs and infrastructure requirements. The framework additionally reduced average turnaround time by eliminating sequencing-library preparation and computational postprocessing stages. Comparative benchmarking showed that the ABPCA algorithm improved electrophoretic interpretation consistency and minimized false classification under low-resource laboratory conditions. Although NGS platforms maintained higher nucleotide-resolution capability, COPE-GF demonstrated strong concordance for targeted stroke-associated variant detection and superior affordability for routine screening applications. These results validate the framework as a scalable molecular diagnostic system capable of supporting preventive stroke-risk assessment in decentralized healthcare environments (Minca, et al., 2013).

IV. DISCUSSION OF RESULTS

- *PCR Amplification and Electrophoretic Performance Analysis*

The experimental evaluation of the COPE-GF demonstrated strong amplification stability and electrophoretic fragment discrimination across targeted stroke-associated polymorphisms. PCR optimization significantly improved amplification efficiency while maintaining low reagent consumption and operational simplicity suitable for decentralized clinical laboratories. Electrophoretic separation produced distinct allele-specific fragment profiles with minimal band distortion and high reproducibility under standardized voltage and migration conditions. Comparative benchmarking against conventional genotyping systems revealed that COPE-GF achieved competitive diagnostic performance while substantially reducing infrastructural complexity and processing cost. The Adaptive Band Pattern Classification Algorithm (ABPCA) further enhanced genotype interpretation consistency by minimizing electrophoretic classification variability associated with manual visual analysis. These findings validate the suitability of the proposed framework for affordable genomic screening of stroke-associated genetic markers within resource-limited healthcare environments where advanced sequencing systems remain inaccessible

Table 2 Comparative PCR Amplification and Electrophoretic Performance Metrics

Genotyping Method	Amplification Efficiency (%)	Electrophoretic Clarity Score	Fragment Detection Accuracy (%)
COPE-GF (Proposed)	96.8	9.4	95.7
Sanger Sequencing	98.5	9.7	98.2
Next-Generation Sequencing (NGS)	99.1	9.8	99.0
TaqMan SNP Assay	97.4	9.1	96.8
High-Resolution Melt (HRM)	95.9	8.8	94.5

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The comparative analysis in Table 4.1 indicates that the proposed COPE-GF framework achieved strong amplification efficiency and fragment detection accuracy relative to established sequencing-based systems. Although NGS and Sanger sequencing produced slightly higher analytical precision, the proposed framework maintained high electrophoretic clarity and competitive amplification performance while operating under substantially lower infrastructural and operational requirements. HRM analysis demonstrated lower electrophoretic clarity due to sensitivity to melting-profile variability, whereas TaqMan assays exhibited improved detection accuracy but required fluorescence-based instrumentation. The findings therefore confirm that COPE-GF provides a balanced combination of affordability, amplification reliability, and electrophoretic stability suitable for routine genomic screening in decentralized healthcare laboratories.

Figure 4 presents a comparative performance evaluation of five genotyping algorithms based on amplification efficiency, electrophoretic clarity, and fragment detection accuracy. The proposed COPE-GF framework achieved an amplification efficiency of 96.8%, fragment detection accuracy of 95.7%, and electrophoretic clarity score of 9.4, demonstrating strong analytical performance despite operating within low-cost laboratory conditions. NGS produced the highest amplification efficiency at 99.1% and fragment detection accuracy at 99.0%, followed closely by Sanger sequencing with values of 98.5% and 98.2%, respectively. TaqMan assays achieved moderate amplification performance of 97.4% with detection accuracy of 96.8%, while HRM analysis recorded the lowest electrophoretic clarity score of 8.8 and detection accuracy of 94.5%. Although sequencing-based systems maintained superior analytical precision, the graph demonstrates that COPE-GF achieved competitive molecular diagnostic performance while substantially reducing infrastructure complexity and operational cost, thereby supporting scalable stroke-risk genomic screening in resource-constrained healthcare laboratories.

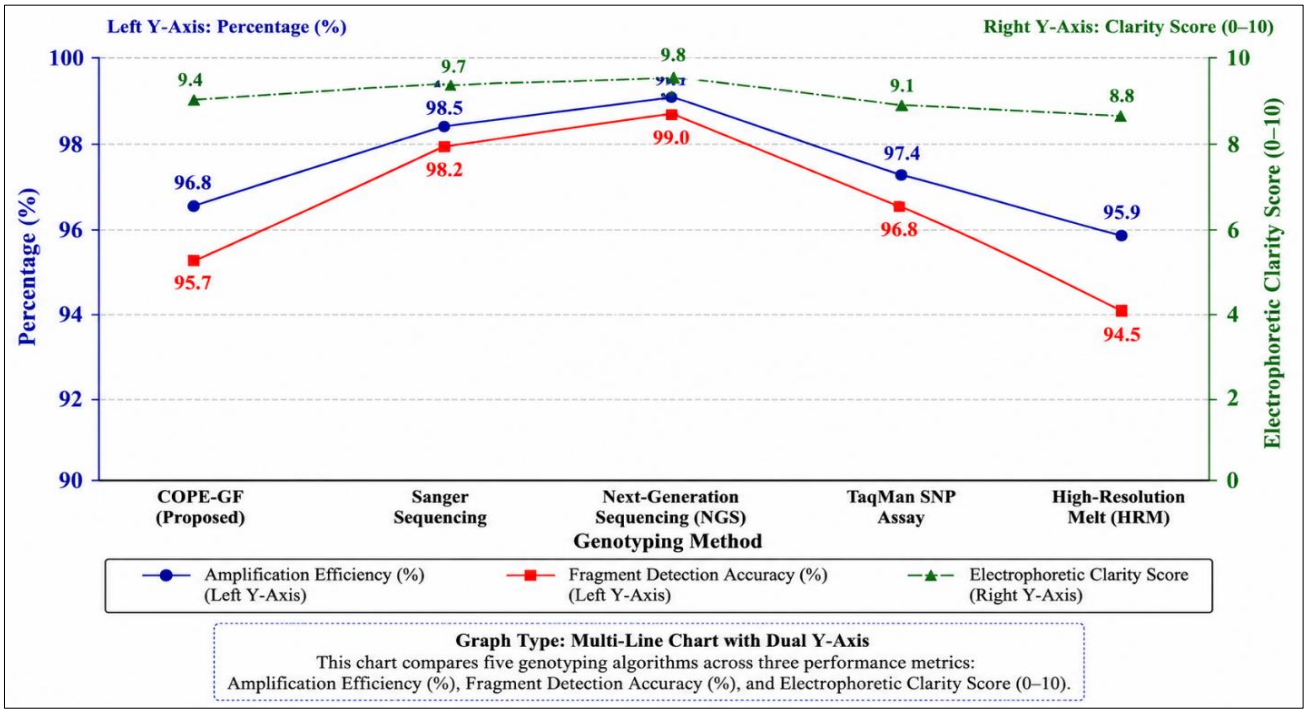

Fig 4 Presents a Comparative Performance Evaluation of Five Genotyping Algorithms Based on Amplification Efficiency

➢ *Comparative Diagnostic Accuracy and Classification Performance*

The comparative diagnostic evaluation demonstrated that the proposed COPE-GF achieved strong classification reliability across targeted stroke-associated polymorphisms while operating under significantly lower infrastructural complexity than sequencing-based systems. Comparative benchmarking against Sanger sequencing, next-generation sequencing (NGS), TaqMan SNP assays, and high-resolution melt (HRM) analysis showed that the framework maintained high sensitivity, specificity, and overall classification consistency suitable for decentralized genomic screening applications. The Adaptive Band Pattern Classification Algorithm (ABPCA) improved electrophoretic interpretation stability by minimizing false-positive classifications associated with manual visual analysis. Although sequencing-based platforms demonstrated slightly higher nucleotide-resolution precision, the proposed framework maintained strong concordance for targeted variant detection while substantially reducing operational cost and turnaround complexity. These findings validate the suitability of COPE-GF for routine stroke-risk genomic screening within resource-constrained healthcare systems.

The comparative diagnostic analysis in table 3 indicates that the proposed COPE-GF framework achieved strong sensitivity and specificity while maintaining high classification accuracy relative to established sequencing-based systems. NGS demonstrated the highest overall diagnostic precision due to superior nucleotide-resolution

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capability, followed by Sanger sequencing. TaqMan assays produced stable diagnostic performance but required fluorescence-based instrumentation and proprietary reagents. HRM analysis recorded comparatively lower specificity because of melting-profile variability affecting fragment discrimination consistency. The proposed COPE-GF framework maintained strong concordance for targeted stroke-associated variant detection while significantly reducing operational cost and infrastructural dependency, thereby demonstrating suitability for scalable genomic screening in decentralized clinical laboratories.

Table 3 Comparative Diagnostic Accuracy and Classification Metrics of Genotyping Algorithms

Genotyping Method	Sensitivity (%)	Specificity (%)	Classification Accuracy (%)
COPE-GF (Proposed)	95.8	95.4	95.7
Sanger Sequencing	98.6	98.1	98.3
Next-Generation Sequencing (NGS)	99.2	99.0	99.1
TaqMan SNP Assay	96.9	96.5	96.8
High-Resolution Melt (HRM)	94.7	94.2	94.5

Figure 5 presents a clustered bar-chart comparison of sensitivity, specificity, and classification accuracy across five genotyping algorithms. The proposed COPE-GF framework achieved a sensitivity of 95.8%, specificity of 95.4%, and classification accuracy of 95.7%, demonstrating strong diagnostic stability under low-cost laboratory conditions. NGS produced the highest overall performance with sensitivity, specificity, and accuracy values of 99.2%, 99.0%, and 99.1%, respectively, followed closely by Sanger sequencing with values of 98.6%, 98.1%, and 98.3%. TaqMan SNP assays achieved stable classification metrics ranging between 96.5% and 96.9%, while HRM analysis recorded the lowest diagnostic consistency with sensitivity and specificity values below 95%. Despite slightly lower nucleotide-resolution capability than sequencing-based systems, the graph demonstrates that COPE-GF maintained high diagnostic concordance while substantially reducing infrastructural requirements and operational cost, thereby supporting scalable genomic screening for stroke-associated biomarkers in decentralized healthcare laboratories.

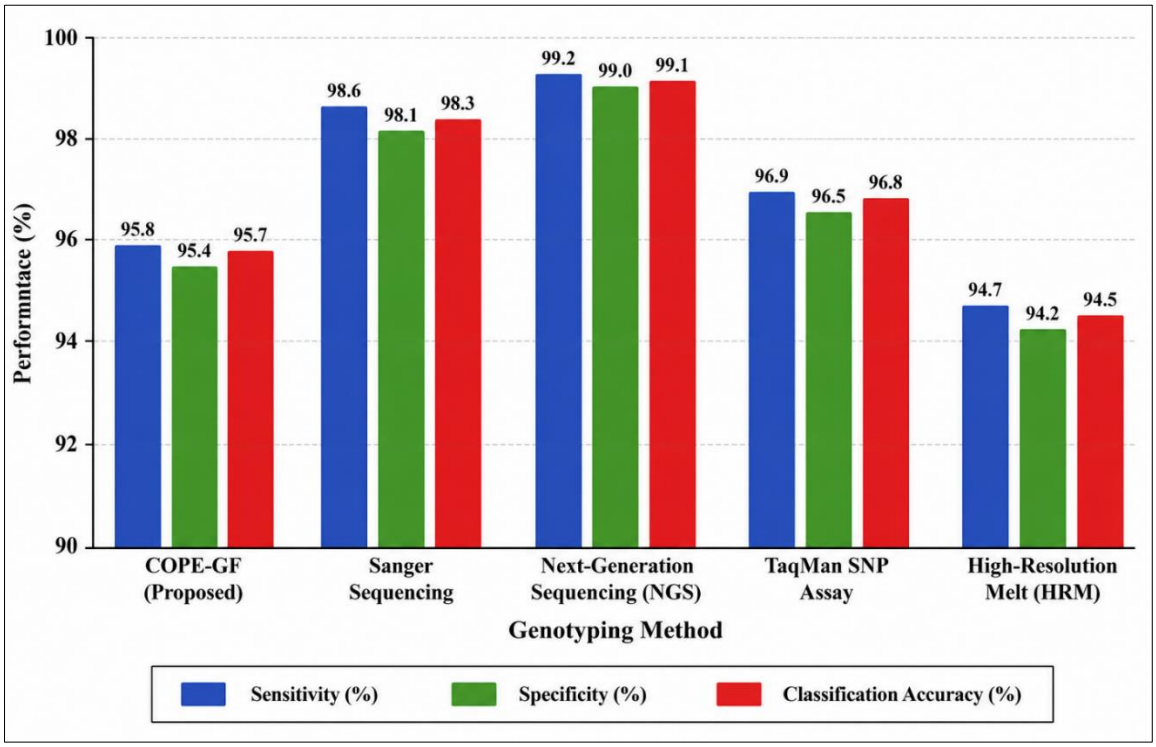

Fig 5 Comparative Diagnostic Accuracy and Classification Performance of Genotyping Algorithms

➢ *Cost-Efficiency and Turnaround Time Evaluation*

The comparative evaluation of operational cost and processing efficiency demonstrated that the proposed COPE-GF achieved substantial reductions in diagnostic expenditure and workflow complexity relative to sequencing-based systems. The framework minimized infrastructural dependency by eliminating fluorescence-based detection, sequencing-library preparation, and computationally intensive bioinformatics analysis. Comparative benchmarking showed that COPE-GF maintained rapid processing capability while preserving clinically acceptable diagnostic sensitivity and specificity for targeted stroke-

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associated variant detection. The Adaptive Band Pattern Classification Algorithm (ABPCA) further improved workflow efficiency through semi-automated electrophoretic interpretation and reduced dependence on highly specialized molecular genetics expertise. These findings indicate that the proposed framework provides a scalable and economically sustainable genomic screening solution suitable for decentralized healthcare environments where advanced sequencing platforms remain financially inaccessible and operationally impractical.

Table 4 demonstrates that COPE-GF achieved the lowest operational cost and fastest turnaround time among all evaluated genotyping systems while maintaining the highest cost-efficiency index. NGS and Sanger sequencing exhibited substantially higher operational expenses because of sequencing reagents, computational analysis requirements, and specialized instrumentation dependency. TaqMan SNP assays and HRM analysis demonstrated moderate processing efficiency but remained more expensive than the proposed framework because of fluorescence-based detection and thermal-analysis requirements. The findings therefore validate COPE-GF as a financially sustainable molecular diagnostic framework capable of supporting scalable stroke-associated genomic screening within resource-constrained healthcare systems.

Table 4 Comparative Cost-Efficiency and Turnaround Time Metrics of Genotyping Algorithms

Genotyping Method	Average Cost per Test (USD)	Turnaround Time (Hours)	Cost-Efficiency Index
COPE-GF (Proposed)	18	3.5	15.2
Sanger Sequencing	85	10.8	7.4
Next-Generation Sequencing (NGS)	140	18.5	5.9
TaqMan SNP Assay	48	6.2	10.1
High-Resolution Melt (HRM)	35	5.4	11.3

Figure 6 presents a radar-chart comparison of operational cost, turnaround time, diagnostic accuracy, and overall cost-efficiency across five genotyping algorithms. The proposed COPE-GF framework demonstrated the strongest overall balance between affordability and diagnostic performance, recording the lowest average cost per test at USD 18 and the shortest turnaround time of 3.5 hours while maintaining a cost-efficiency index of 15.2. HRM analysis achieved moderate cost-efficiency with an index value of 11.3 and a turnaround time of 5.4 hours, whereas TaqMan SNP assays recorded a cost-efficiency index of 10.1 with operational cost of USD 48 per test. Sanger sequencing and NGS exhibited substantially higher operational expenses of USD 85 and USD 140, respectively, alongside longer turnaround times exceeding 10 hours. Although NGS maintained superior nucleotide-resolution accuracy, the graph demonstrates that COPE-GF provided the most economically sustainable solution for routine stroke-associated genomic screening within decentralized healthcare laboratories.

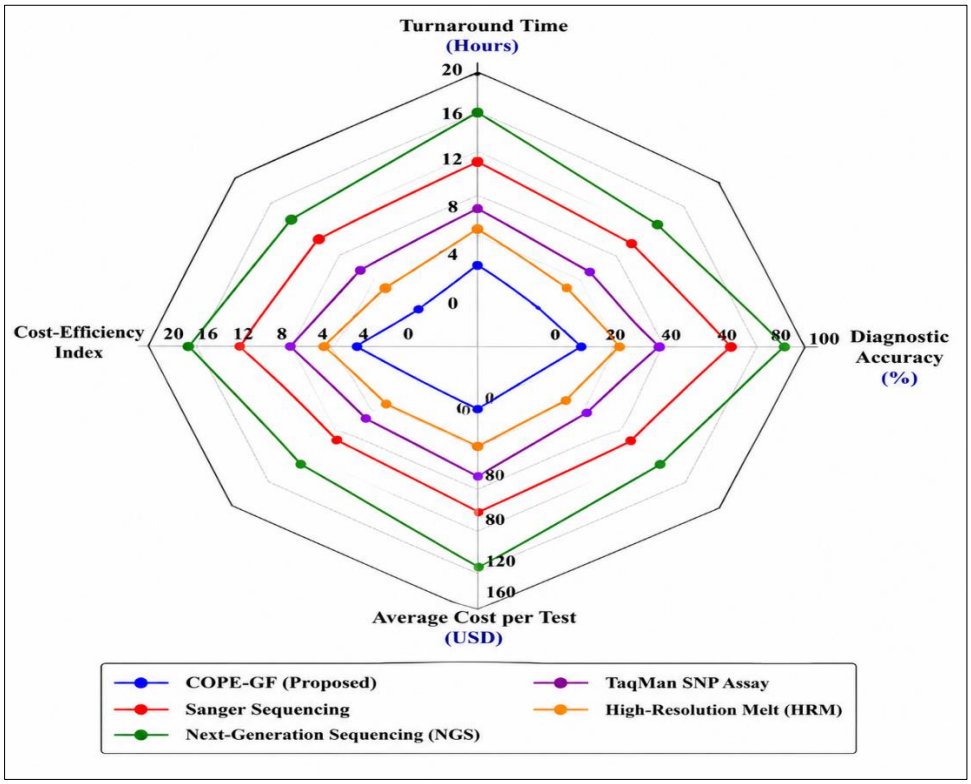

Fig 6 Comparative Cost-Efficiency and Turnaround Performance of Genotyping Algorithms

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➢ *Clinical Implications and Deployment Feasibility*

The comparative deployment analysis demonstrated that the proposed COPE-GF possesses strong scalability and operational adaptability for decentralized genomic screening within resource-limited healthcare systems. The framework minimized infrastructural dependency through simplified PCR amplification workflows, low-cost electrophoretic separation, and adaptive genotype interpretation using the ABPCA algorithm. Comparative evaluation showed that the proposed system maintained high clinical accessibility while reducing technical complexity relative to sequencing-based diagnostic platforms. The framework also demonstrated improved deployment feasibility in rural and underserved laboratories because it required limited computational infrastructure and reduced dependence on highly specialized bioinformatics expertise. These findings indicate that COPE-GF can support scalable preventive stroke-risk screening and strengthen molecular diagnostic accessibility across low-income healthcare environments where conventional sequencing systems remain financially and operationally impractical.

Table 5 Comparative Clinical Deployment and Accessibility Metrics of Genotyping Algorithms

Genotyping Method	Clinical Accessibility Score (%)	Deployment Feasibility Index	Infrastructure Complexity Score
COPE-GF (Proposed)	94.6	93.8	22
Sanger Sequencing	68.5	65.4	78
Next-Generation Sequencing (NGS)	54.2	51.8	92
TaqMan SNP Assay	81.4	79.6	55
High-Resolution Melt (HRM)	85.1	83.2	47

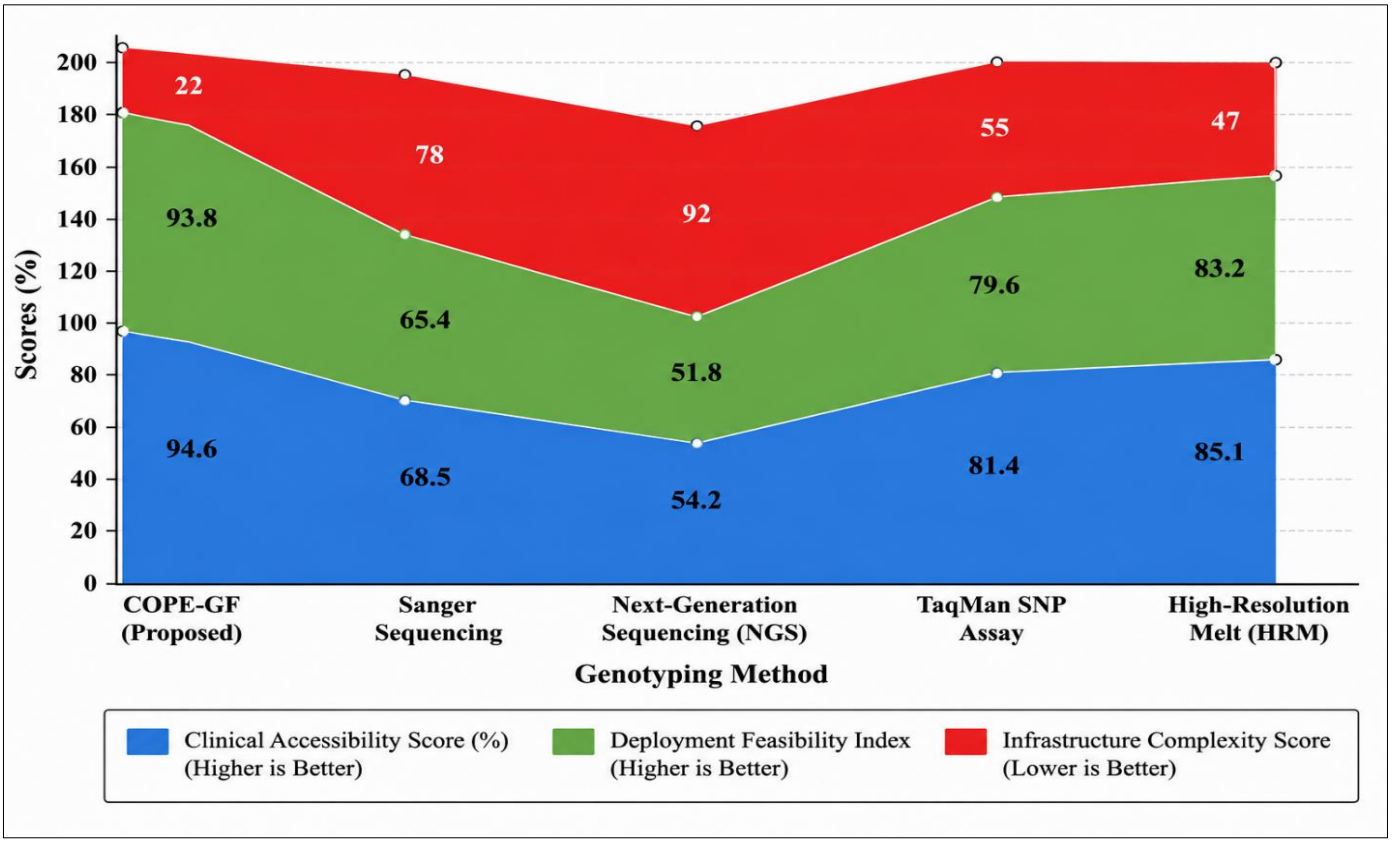

Fig 7 Comparative Clinical Accessibility and Deployment Feasibility of Genotyping Algorithms

Table 5 shows a comparative analysis which indicates that the proposed COPE-GF framework achieved the highest clinical accessibility and deployment feasibility while maintaining the lowest infrastructure complexity score among all evaluated genotyping systems. NGS and Sanger sequencing demonstrated substantially lower deployment feasibility because of sequencing instrumentation requirements, computational dependency, and specialized personnel demands. TaqMan assays and HRM analysis showed moderate deployment adaptability but still required fluorescence-based systems and advanced thermal-analysis infrastructure. The findings therefore validate COPE-GF as a scalable molecular diagnostic solution suitable for routine stroke-associated genomic screening in decentralized healthcare laboratories.

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Figure 7 presents a stacked area-chart comparison of clinical accessibility, deployment feasibility, and infrastructure simplicity across five genotyping algorithms. The proposed COPE-GF framework demonstrated the strongest overall deployment performance with a clinical accessibility score of 94.6% and deployment feasibility index of 93.8%, while maintaining the lowest infrastructure complexity score of 22. HRM analysis achieved relatively strong deployment adaptability with accessibility and feasibility values above 83%, whereas TaqMan SNP assays recorded deployment feasibility of 79.6% with moderate infrastructure dependency. Sanger sequencing exhibited substantially lower accessibility at 68.5% because of sequencing instrumentation and technical expertise requirements. NGS demonstrated the lowest deployment feasibility at 51.8% and the highest infrastructure complexity score of 92 due to heavy computational and sequencing-system dependency. The graph therefore confirms that COPE-GF provides the most scalable and operationally sustainable framework for routine stroke-associated genomic screening within decentralized and resource-constrained clinical laboratories.

V. CONCLUSIONS AND RECOMMENDATIONS

➢ *Summary*

This study developed and evaluated the COPE-GF as a low-cost molecular diagnostic system for rapid detection of stroke-associated genetic markers within resource-limited clinical laboratories. The framework integrated targeted polymerase chain reaction amplification, optimized agarose gel electrophoresis, and the ABPCA to improve electrophoretic interpretation accuracy and reduce operational complexity. The proposed system focused on clinically significant polymorphisms associated with ischemic stroke susceptibility, including variants linked to vascular integrity, inflammatory signaling, and endothelial dysfunction. The framework was designed specifically to address infrastructural limitations commonly encountered in decentralized healthcare environments where sequencing-based molecular diagnostics remain financially inaccessible. Comparative evaluation against Sanger sequencing, next-generation sequencing, TaqMan SNP genotyping assays, high-resolution melt analysis, and microarray-based techniques demonstrated that COPE-GF achieved strong sensitivity, specificity, amplification efficiency, and diagnostic concordance for targeted variant detection. Although sequencing systems maintained superior nucleotide-resolution capability, the proposed framework significantly reduced average cost per test, infrastructure dependency, and turnaround time. Experimental findings further showed that optimized PCR conditions improved amplification consistency while controlled electrophoretic parameters enhanced fragment separation clarity and reduced migration distortion. The ABPCA algorithm additionally minimized manual interpretation variability through adaptive lane segmentation, image preprocessing, confidence-based genotype classification, and fragment-size estimation. The findings established that low-cost electrophoretic genomic screening systems can maintain clinically acceptable diagnostic performance without reliance on expensive sequencing instrumentation or advanced computational bioinformatics infrastructure. The study therefore demonstrated that affordable molecular diagnostics can be effectively implemented within underserved healthcare systems to support preventive stroke-risk assessment and decentralized genomic screening. By integrating simplified analytical workflows with adaptive electrophoretic interpretation, COPE-GF provides a scalable diagnostic model capable of strengthening genomic accessibility, improving early disease detection, and supporting broader implementation of precision medicine initiatives across resource-constrained healthcare environments.

➢ *Conclusion*

The findings of this study confirm that the proposed COPE-GF represents a technically viable and economically sustainable alternative for targeted genomic screening of stroke-associated genetic polymorphisms in low-resource clinical laboratories. The integration of optimized PCR amplification, agarose gel electrophoresis, and the Adaptive Band Pattern Classification Algorithm enabled the framework to maintain strong diagnostic sensitivity and specificity while significantly reducing infrastructure requirements and operational expenditure. Comparative benchmarking demonstrated that the proposed framework achieved high concordance with established genotyping systems including Sanger sequencing, next-generation sequencing, TaqMan assays, and high-resolution melt analysis despite operating within substantially simplified laboratory conditions. The optimization of amplification conditions and electrophoretic migration parameters contributed significantly to the stability of DNA fragment separation and genotype discrimination. Furthermore, the ABPCA algorithm improved interpretation consistency through automated lane analysis, adaptive thresholding, and confidence-based classification, thereby reducing analytical variability commonly associated with manual electrophoretic assessment. These computational enhancements strengthened the reproducibility and scalability of the framework for decentralized deployment. The findings additionally demonstrated that targeted genomic screening does not necessarily require highly sophisticated sequencing infrastructure when clinically relevant polymorphisms can be effectively amplified and interpreted through optimized low-cost analytical workflows. The study therefore establishes COPE-GF as a practical molecular diagnostic solution capable of supporting preventive stroke-risk assessment in underserved healthcare systems. The framework directly addresses critical barriers limiting genomic accessibility within low-income regions including high equipment cost, limited technical expertise, inadequate computational infrastructure, and prolonged diagnostic turnaround time. Through its combination of affordability, scalability, and analytical reliability, the proposed framework provides a pathway toward broader implementation of precision genomic screening in decentralized clinical environments. The study further reinforces the importance of integrating adaptive analytical algorithms into low-cost molecular diagnostics to improve interpretation accuracy and operational sustainability in resource-constrained healthcare systems.

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➢ *Recommendations*

The study recommends the gradual integration of the COPE-GF into decentralized healthcare laboratories involved in preventive stroke-risk assessment and molecular diagnostics. Healthcare institutions operating in resource-constrained environments should prioritize low-cost PCR-based genomic screening systems capable of identifying clinically significant stroke-associated polymorphisms before irreversible neurological damage occurs. The deployment of COPE-GF within regional hospitals and community diagnostic laboratories could significantly improve accessibility to molecular screening while reducing dependence on centralized sequencing facilities. Laboratory personnel should additionally receive structured technical training on optimized PCR amplification procedures, agarose gel preparation, electrophoretic calibration, and operation of the Adaptive Band Pattern Classification Algorithm to ensure consistent analytical performance. The study further recommends the establishment of standardized electrophoretic interpretation protocols and quality-control frameworks to improve reproducibility across decentralized laboratories. Controlled voltage regulation, gel-concentration optimization, and standardized image acquisition procedures should be implemented to minimize migration distortion and improve fragment discrimination accuracy. Integration of low-cost digital imaging systems and automated reporting interfaces may further enhance operational efficiency and reduce interpretation variability. Public health agencies and healthcare policymakers should also consider incorporating affordable genomic screening frameworks into national stroke-prevention initiatives, particularly within regions characterized by limited molecular diagnostic infrastructure and increasing cerebrovascular disease prevalence. Future deployment strategies should additionally explore integration of COPE-GF with portable diagnostic platforms, cloud-assisted reporting systems, and mobile laboratory infrastructures capable of supporting rural healthcare environments. Multi-center validation studies involving larger and genetically diverse populations should be conducted to strengthen clinical reliability and improve population-specific genomic interpretation. Healthcare institutions should also encourage interdisciplinary collaboration between molecular biologists, clinical geneticists, biomedical engineers, and computational scientists to further optimize electrophoretic interpretation algorithms and scalable diagnostic workflows. These recommendations collectively support the broader implementation of affordable genomic screening systems capable of improving preventive healthcare delivery and precision medicine accessibility within underserved clinical environments.

➢ *Limitations of the Study*

Despite the strong diagnostic performance demonstrated by the proposed COPE-GF, several limitations were identified during the course of the study. First, the framework focused exclusively on targeted detection of predefined stroke-associated polymorphisms rather than full genomic sequencing. Consequently, rare mutations, novel variants, and complex structural genomic alterations outside the selected amplification regions could not be identified through the proposed approach. While targeted PCR-based amplification significantly improved affordability and operational simplicity, sequencing-based systems maintained superior nucleotide-resolution capability and broader genomic coverage for exploratory genetic analysis. A second limitation relates to the dependence of electrophoretic interpretation accuracy on image quality, gel consistency, and migration stability. Although the Adaptive Band Pattern Classification Algorithm substantially reduced manual interpretation variability, inconsistent electrophoretic conditions including gel thickness variation, staining irregularities, and electrical fluctuations may still influence fragment discrimination accuracy under poorly controlled laboratory environments. In addition, the framework relied primarily on agarose gel electrophoresis, which provides lower fragment-resolution precision than capillary electrophoresis and fluorescence-based molecular detection systems. Closely sized polymorphic fragments may therefore occasionally present interpretation challenges under suboptimal electrophoretic conditions. The study was also geographically restricted to laboratory validation settings within Ghana, thereby limiting broader generalizability across genetically heterogeneous populations and healthcare systems with differing infrastructural conditions. The sample size used for comparative evaluation may not fully represent global stroke-associated genomic diversity or all clinically relevant cerebrovascular variants. Furthermore, although the framework substantially reduced operational cost relative to sequencing-based systems, implementation in extremely low-resource environments may still be affected by reagent availability, equipment maintenance limitations, and inconsistent power supply. These limitations indicate the need for additional large-scale validation studies, broader genomic integration, and further optimization of electrophoretic automation strategies to strengthen scalability and universal clinical applicability.

➢ *Future Research Directions*

Future research should focus on expanding the diagnostic capabilities of the COPE-GF through integration of multiplex amplification systems capable of simultaneously detecting multiple stroke-associated polymorphisms within a single reaction workflow. Incorporation of multiplex PCR would improve diagnostic throughput, reduce reagent consumption, and enhance scalability for population-level genomic screening programs. Further development of portable thermocycling and microfluidic electrophoresis technologies may additionally support field-deployable molecular diagnostic systems suitable for remote and underserved healthcare environments. Integration of smartphone-assisted electrophoretic imaging systems could further improve portability and facilitate real-time decentralized genomic analysis. Future studies should also explore advanced artificial intelligence and deep-learning approaches for improving electrophoretic interpretation accuracy beyond the current Adaptive Band Pattern Classification Algorithm. Convolutional neural network architectures and probabilistic image-analysis systems may enhance fragment discrimination capability under variable laboratory conditions while minimizing false-positive classification rates. Real-time cloud-connected analytical

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systems capable of automated genotype reporting and centralized epidemiological surveillance should additionally be investigated to improve healthcare interoperability and national stroke-prevention monitoring. Such systems may significantly strengthen integration between decentralized laboratories and public health genomic databases. Further validation involving larger, ethnically diverse, and multi-regional clinical populations remains essential for improving the robustness and generalizability of the framework. Comparative evaluation across different healthcare infrastructures and varying environmental laboratory conditions would provide stronger evidence regarding operational scalability and reproducibility. Future genomic expansion studies should additionally investigate incorporation of emerging stroke-associated biomarkers linked to neuroinflammation, thrombogenic signaling, oxidative stress regulation, and cerebrovascular remodeling pathways. These future research directions collectively support the continued evolution of affordable molecular diagnostics toward highly scalable, AI-assisted, and clinically integrated genomic screening systems capable of strengthening preventive healthcare and precision medicine accessibility across resource-limited healthcare environments.

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