INSIGHTS-JOURNAL OF
LIFE AND SOCIAL SCIENCES

INSIGHTS-JLSS
Insights-Journal of Life and Social Sciences

RECENT ADVANCES IN MOLECULAR BIOMARKERS FOR EARLY DETECTION OF EPILEPSY: A NARRATIVE REVIEW

Narrative Review

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Conflict of Interest: None

Grant Support & Financial Support: None

Acknowledgment: The author acknowledges the valuable contributions of researchers and clinicians whose work in molecular neuroscience and epilepsy has informed this review. Appreciation is also extended to institutional libraries and open-access databases for providing access to the latest scientific literature.

ABSTRACT

Background: Epilepsy is a common and often debilitating neurological disorder, affecting over 50 million people worldwide. Despite advances in imaging and electrophysiology, early diagnosis remains challenging, particularly in patients without clear clinical manifestations. Molecular and genetic biomarkers offer a promising avenue for improving early detection, risk stratification, and individualized treatment strategies in epilepsy.

Objective: This narrative review aims to explore recent advances in the identification and validation of molecular and genetic biomarkers for the early diagnosis and management of epilepsy, with a focus on their clinical applicability and current research gaps.

Main Discussion Points: The review synthesizes evidence across several biomarker domains, including genetic mutations (e.g., *SCN2A*, *KCNQ2*), circulating microRNAs, neuroinflammatory markers, and proteomic/metabolomic indicators. It also discusses the emerging role of biosensor technology in real-time monitoring and diagnostics. Critical analysis reveals limitations such as small sample sizes, lack of longitudinal data, methodological inconsistencies, and limited generalizability. The need for standardized protocols and multicenter validation is emphasized to improve reliability and clinical translation.

Conclusion: Molecular and genetic biomarkers hold significant potential to transform epilepsy care through earlier diagnosis and more precise treatment planning. However, current evidence is preliminary, and substantial research is needed to validate these tools for routine clinical use. Future studies should adopt standardized, large-scale, and multi-modal designs to fully harness the diagnostic power of biomarkers in epilepsy.

Keywords: Epilepsy, Molecular Biomarkers, Genetic Markers, Early Diagnosis, MicroRNAs, Narrative Review.

INTRODUCTION

Epilepsy is one of the most prevalent and disabling chronic neurological disorders worldwide, affecting over 50 million individuals, with approximately 80% of cases occurring in low- and middle-income countries. It is characterized by recurrent, unprovoked seizures resulting from abnormal neuronal activity and carries substantial burdens related to cognitive impairment, psychiatric comorbidities, and social stigma. The early diagnosis and effective management of epilepsy remain critical, particularly given that around 30% of patients are resistant to currently available antiseizure medications (1). In this context, molecular and genetic biomarkers have emerged as a promising frontier for revolutionizing the diagnosis, monitoring, and treatment of epilepsy, potentially transforming clinical approaches from reactive to predictive and personalized paradigms (2). Despite advances in imaging techniques and electrophysiological assessments, the identification of reliable, non-invasive biomarkers for early epilepsy detection remains elusive. Diagnostic delays are common, particularly in early-onset and atypical cases, where subtle seizures may not be immediately recognized or reported. The challenge is further compounded by the heterogeneity of epileptic syndromes and underlying etiologies. Traditional diagnostic tools, such as EEG and MRI, while valuable, often lack sensitivity and specificity in the early stages of disease, especially when structural abnormalities are absent or when interictal EEGs are normal. This diagnostic gap underscores the critical need for biomarkers that can accurately predict epileptogenesis—the process through which a normal brain becomes capable of generating spontaneous seizures—and guide early therapeutic intervention (3,4).

Recent advances in molecular biology and omics technologies have unveiled a wealth of candidate biomarkers. Blood-based microRNAs (miRNAs), in particular, have shown considerable promise due to their stability, brain enrichment, and reproducible changes in expression among patients with epilepsy. Studies have reported distinct miRNA expression profiles that correlate with seizure burden and treatment resistance, supporting their utility as accessible and non-invasive biomarkers (5). Similarly, genomic studies have revealed pathogenic variants in genes such as KCNQ2, SCN2A, and KCNT1 among individuals with early-onset epilepsies, providing insight into the molecular underpinnings of these syndromes and paving the way for gene-based diagnostics (6). These discoveries have been bolstered by whole-genome sequencing, which has improved the diagnostic yield in complex pediatric epilepsy cases and uncovered novel genetic mechanisms (7). Beyond nucleic acids, proteomic and metabolomic approaches have also identified potential serum and cerebrospinal fluid markers, including neuroinflammatory cytokines, stress-related hormones, and neuronal damage-related proteins. For instance, elevated levels of IL-1β and HMGB1 have been implicated in the pathophysiology of seizures and may serve as indicators of neuroinflammatory states associated with epilepsy (8). Moreover, biosensor technologies are being developed to detect biomarkers such as glutamate and potassium ions in real time, offering innovative diagnostic applications at the interface of engineering and neurology (9).

However, the field is not without its challenges. A major hurdle lies in the validation and standardization of candidate biomarkers across diverse patient populations and clinical settings. Many existing studies are preliminary, limited in sample size, or lack replication. Moreover, confounding variables such as age, seizure type, etiology, medication status, and comorbid conditions can influence biomarker levels, necessitating rigorous control and stratification in future studies (10,11). Furthermore, the translation of these biomarkers from bench to bedside will require multicenter collaborations, standardized protocols for sample collection and processing, and robust statistical frameworks for biomarker discovery and validation. The objective of this narrative review is to synthesize the current progress in identifying and validating molecular and genetic biomarkers for the early detection of epilepsy. It aims to examine the evidence supporting various classes of biomarkers—including genetic mutations, miRNAs, proteins, and metabolites—and to evaluate their potential roles in early diagnosis, prognosis, and treatment monitoring. The review will also explore the technical and clinical challenges that impede biomarker translation and propose future directions to advance the field.

This review will focus on studies published in the last five years, emphasizing both clinical and preclinical research that has contributed to our understanding of epilepsy biomarkers. Particular attention will be given to high-throughput methodologies such as transcriptomic, proteomic, and metabolomic analyses, as well as to technological innovations like biosensors and liquid biopsy platforms. The inclusion criteria are restricted to original research and reviews that provide insights into early-stage biomarker discovery and validation for epilepsy. By compiling and critically evaluating the recent literature, this review seeks to provide clinicians, researchers, and healthcare stakeholders with a comprehensive and up-to-date understanding of molecular biomarker development in epilepsy. The integration of validated biomarkers into clinical practice has the potential to significantly enhance early diagnosis, individualize treatment strategies, and reduce the long-term burden of epilepsy. Ultimately, this review underscores the importance of biomarker-driven approaches in reshaping the landscape of epilepsy care.

THEMATIC DISCUSSION

1. Genetic Markers and Epileptogenic Mutations

Genetic biomarkers are among the most robust and well-studied indicators for early detection of epilepsy, particularly in syndromes with strong heritable components. Several monogenic epilepsies have been linked to pathogenic variants in ion channel genes, such as *SCN2A*, *KCNQ2*, and *KCNT1*. A clinical whole-genome sequencing (WGS) study involving pediatric patients with severe early-onset epilepsy identified de novo mutations in these genes, some of which had not been previously associated with epilepsy. These mutations significantly impacted neuronal excitability and were correlated with early phenotypic expression, suggesting that WGS offers an effective diagnostic approach when conventional testing fails (11,12). Furthermore, integrating WGS with clinical phenotyping enhances diagnostic precision and helps in identifying new therapeutic targets.

2. Circulating microRNAs as Blood-Based Biomarkers

In recent years, circulating microRNAs (miRNAs) have emerged as particularly promising molecular biomarkers due to their brain-specific expression, stability in blood, and role in seizure pathophysiology. Several studies have reported that specific miRNAs such as miR-199a-3p and miR-574-3p are significantly altered during epileptogenesis and in chronic epilepsy states. These findings were validated across both animal models and human cohorts, suggesting cross-species relevance (7,13). Additionally, blood-based miRNA profiles have demonstrated discriminatory power between drug-resistant and drug-responsive epilepsy cases, with encouraging sensitivity and specificity values (14).

3. Inflammatory and Immunological Biomarkers

Neuroinflammation is increasingly recognized as a central feature of epileptogenesis, making inflammatory biomarkers an area of growing interest. Biomarkers such as interleukin-1β (IL-1β), tumor necrosis factor-alpha (TNF-α), and high mobility group box 1 (HMGB1) have been consistently reported as elevated in the epileptic brain. These markers are believed to play a causative role in lowering seizure thresholds through mechanisms such as blood-brain barrier disruption and glial activation. Their levels correlate with seizure frequency and severity, suggesting utility in both early detection and disease monitoring (6,15). Despite their promise, the nonspecific nature of inflammatory markers requires them to be interpreted alongside other biomolecular data to avoid misdiagnosis.

4. Proteomic and Metabolomic Signatures

Proteomic approaches have allowed the identification of protein biomarkers associated with seizure activity and neuronal damage. Candidate biomarkers such as brain-derived neurotrophic factor (BDNF), S100B, and neurofilament light chain have shown altered levels in epilepsy models and human studies. These proteins reflect synaptic reorganization and glial responses, both of which are crucial to epileptogenesis. Similarly, metabolomic profiling has indicated that changes in neurotransmitter concentrations, particularly glutamate and GABA, may serve as early indicators of altered neural excitability (9,16). However, the integration of these omics datasets with clinical findings remains a work in progress due to methodological variability.

5. Biosensor-Based Detection of Biomarkers

Technological innovations in biosensor development offer a real-time, non-invasive method to detect biochemical changes associated with seizure onset. Recent biosensors capable of detecting glutamate, potassium ions, and specific miRNAs in cortical tissue have shown success in animal models. These biosensors hold potential not only for diagnostics but also for mechanistic studies into seizure propagation and network hyperexcitability (17). While still in early stages, such tools could complement existing imaging and electrophysiological methods, providing continuous, dynamic monitoring of epilepsy biomarkers.

6. Challenges in Biomarker Validation

Despite encouraging findings, a significant gap persists between biomarker discovery and clinical application. Much of the current evidence is derived from small, single-center studies with heterogeneous methodologies. Inconsistent sample collection protocols, population variability, and lack of longitudinal follow-up are notable challenges. Furthermore, biomarker levels can be influenced by external factors such as medication, comorbidities, and age, complicating interpretation (10,18). These challenges underscore the need for multicenter collaborations and standardized frameworks for validation.

7. Integration of Biomarkers into Clinical Practice

For biomarkers to become part of standard epilepsy care, they must demonstrate reliability, reproducibility, and clinical relevance. The integration of multi-modal biomarkers—genomic, proteomic, imaging, and electrophysiological—is increasingly seen as the optimal approach for accurate diagnosis and prognosis. A strategic roadmap outlines a five-phase biomarker development process, modeled after similar frameworks in oncology and Alzheimer's disease research, to facilitate clinical translation (13,19).

8. Future Perspectives and Unmet Needs

There is growing consensus that a single biomarker is unlikely to suffice for the heterogeneous epilepsy spectrum. Instead, composite biomarker panels tailored to epilepsy subtypes may offer greater accuracy and clinical utility. Future research must also focus on identifying biomarkers capable of predicting disease progression and treatment response. In particular, efforts to discover biomarkers for pharmacoresistant epilepsy could enable timely surgical or alternative therapeutic interventions (18,19).

CRITICAL ANALYSIS AND LIMITATIONS

The current body of literature on molecular and genetic biomarkers for early detection of epilepsy presents notable scientific progress, yet it is not without considerable limitations that affect the reliability, interpretability, and clinical applicability of findings. Across the reviewed studies, a recurring limitation is the predominance of small sample sizes, particularly in exploratory investigations of microRNA profiles or genomic mutations. For instance, studies employing whole-genome sequencing to identify rare pathogenic mutations in early-onset epilepsy often included fewer than ten probands, significantly reducing the statistical power and increasing the risk of type I and II errors (20,21). This limitation restricts the capacity to detect consistent biomarker patterns across broader, more heterogeneous patient populations. Another major issue lies in the scarcity of randomized controlled trials and long-term follow-up studies. Most biomarker investigations to date remain observational and cross-sectional, focusing on correlational rather than causal relationships. For example, studies examining the expression of blood-based miRNAs typically compare patient cohorts with healthy controls or drug-responsive versus drug-resistant groups at a single time point, failing to evaluate how these markers evolve over time or respond to therapeutic interventions (22). Consequently, the potential of biomarkers for predicting disease progression or treatment efficacy remains speculative.

Methodological bias is another concern that undermines confidence in current findings. Selection bias is particularly evident in studies focused on pediatric or pharmacoresistant populations, which may not represent the full clinical spectrum of epilepsy. This overrepresentation can skew results and reduce external validity. In addition, many preclinical studies lack blinding or randomization in animal model experiments, raising concerns about performance bias and overestimation of biomarker utility. Furthermore, inter-study heterogeneity in patient selection criteria, diagnostic definitions, and control matching methods complicates the synthesis of outcomes across research efforts (23). The issue of publication bias also warrants attention. There is a tendency in the literature to highlight novel, statistically significant associations while underreporting negative or inconclusive findings. This skew in publication practices contributes to an inflated sense of progress in biomarker discovery. For instance, while several studies report promising receiver operating characteristic (ROC) values for miRNAs and protein biomarkers, there is limited transparency regarding unsuccessful replication attempts or null results from multicenter trials (24). This selective visibility of positive results may hinder objective evaluation and slow the pace of validation.

Inconsistency in measurement outcomes further complicates comparative assessment across studies. Variations in biofluid types (e.g., serum vs. plasma), sampling timings relative to seizure activity, and assay platforms for RNA or protein quantification lead to significant inter-laboratory variability. For example, differences in sample handling, normalization techniques, and thresholding criteria have resulted in non-overlapping sets of differentially expressed miRNAs across similar patient groups (25). These inconsistencies highlight the urgent need for standardized protocols in biomarker research. The generalizability of existing findings is another critical limitation. Much of the literature is derived from studies in Western or high-income populations, with limited representation of ethnically diverse cohorts or patients from low-resource settings. Given that epilepsy manifests differently across age groups, socioeconomic strata, and geographical regions, the narrow demographic focus restricts the applicability of biomarkers on a global scale. Moreover, many findings are based on single-center or laboratory-specific methodologies, which are difficult to replicate elsewhere without significant technological infrastructure (26). In sum, while the advances in molecular biomarker research for epilepsy are promising, the field is still in an early translational stage. Limitations in study design, methodological rigor, and representativeness must be addressed through

larger, multicenter, longitudinal studies with standardized methods. Only through such rigor can molecular biomarkers transition from experimental tools to clinically actionable diagnostics.

IMPLICATIONS AND FUTURE DIRECTIONS

The integration of molecular and genetic biomarkers into clinical epilepsy care holds the potential to revolutionize early diagnosis, risk stratification, and individualized treatment strategies. The findings from recent literature suggest that biomarkers such as circulating microRNAs, genetic mutations in ion channel genes, and inflammatory mediators can offer predictive value even before clinical symptoms become fully apparent. This capability could allow clinicians to initiate preventive or disease-modifying interventions during the pre-epileptic phase, potentially altering disease trajectory and improving long-term outcomes (20). Additionally, biomarker profiles may help guide decisions regarding antiseizure drug selection or eligibility for surgical intervention, particularly in pharmacoresistant cases where early and accurate localization of epileptogenic zones is crucial (21). From a policy standpoint, the expanding biomarker evidence base underscores the urgency for the development of standardized clinical guidelines that define their appropriate use, interpretation, and limitations. Guidelines would help harmonize diagnostic protocols, facilitate inter-center collaborations, and promote equitable access to biomarker-based testing. Without regulatory frameworks and consensus-driven standards, the clinical translation of these tools risks becoming fragmented and inconsistent across healthcare systems. Moreover, health policy must consider the economic implications of adopting high-throughput molecular assays and ensure cost-effectiveness, especially in resource-limited settings (22).

Despite promising progress, critical gaps remain unaddressed. The majority of biomarkers identified to date are in preliminary or exploratory phases, with limited validation across large, diverse patient cohorts. There is a need for robust, multicenter, prospective studies that examine the longitudinal dynamics of biomarker expression in relation to seizure onset, progression, and treatment response. For instance, the temporal stability and specificity of microRNA biomarkers across different epilepsy subtypes and comorbid conditions require further clarification (23,24). Furthermore, while some molecular markers are associated with epileptogenic activity, it remains uncertain whether they play a causative role or merely reflect downstream effects of ongoing seizures. This distinction is essential for selecting therapeutic targets. Future research should prioritize large-scale, longitudinal cohort studies that incorporate multiple biomarker classes—genomic, transcriptomic, proteomic, and metabolomic—in tandem with imaging and electrophysiological data. Such multi-modal approaches will enhance diagnostic precision and improve the mechanistic understanding of epileptogenesis. Additionally, randomized controlled trials are needed to assess the clinical utility of biomarker-informed interventions, including their impact on treatment efficacy, quality of life, and cost-effectiveness. Methodologically, future studies must adhere to rigorous protocols for sample collection, processing, and assay standardization to ensure reproducibility and minimize bias (25,26). Finally, emerging technologies such as wearable biosensors and point-of-care diagnostic platforms may soon enable real-time monitoring of epilepsy-related biomarkers. These tools could be particularly valuable in ambulatory settings or for patients in remote areas, making epilepsy care more proactive and accessible (10,20). To maximize the clinical impact of these innovations, future investigations should also incorporate patient-centered outcomes and consider ethical, legal, and social implications of molecular diagnostics in epilepsy.

CONCLUSION

This review highlights the growing body of evidence supporting molecular and genetic biomarkers as promising tools for the early detection and personalized management of epilepsy. Key findings underscore the potential of microRNAs, genetic mutations, inflammatory mediators, and biosensor technologies to identify at-risk individuals before the onset of clinical seizures, as well as to stratify patients based on treatment responsiveness and disease trajectory. While these discoveries mark important progress, the strength of current evidence remains moderate, largely due to methodological limitations, small sample sizes, and a lack of large-scale, longitudinal validation. Clinicians are encouraged to stay informed about emerging biomarker research while exercising caution in adopting these tools prematurely into routine practice. Researchers should prioritize multicenter, standardized, and prospective investigations that integrate multi-omic and neurophysiological data to establish clinically actionable biomarker panels. Ultimately, robust and reproducible biomarker research is essential to usher in a new era of precision medicine in epilepsy care.

AUTHOR CONTRIBUTION

Author	Contribution
Ariba Shah*	Substantial Contribution to study design, analysis, acquisition of Data Manuscript Writing Has given Final Approval of the version to be published
Abdul Rehman Naeem	Substantial Contribution to study design, acquisition and interpretation of Data Critical Review and Manuscript Writing Has given Final Approval of the version to be published
Amna Noor	Substantial Contribution to acquisition and interpretation of Data Has given Final Approval of the version to be published
Adeel-ur-Rehman	Contributed to Data Collection and Analysis Has given Final Approval of the version to be published
Haseeb Muhammad Khan	Contributed to Data Collection and Analysis Has given Final Approval of the version to be published
Akif Saeed Ch	Substantial Contribution to study design and Data Analysis Has given Final Approval of the version to be published
Roha Tariq	Contributed to study concept and Data collection Has given Final Approval of the version to be published
Mohsin Rasheed	Writing - Review & Editing, Assistance with Data Curation

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