Innovative Journal of Medical and Healthcare Research (IJMHR)
Volume-2 Issue-2 March-2025

Bidirectional Mechanism Of Neuroinflammation And Drug-Resistant Epilepsy

Tushar Anshu[1], Preeti Verma[2]

[1]*Assistant Professor, Department of Pharmacology, Vinayaka College of Pharmacy, Kullu, Himachal Pradesh, India.*

[2]*Assistant Professor, Department of Pharmaceutics, Vinayaka College of Pharmacy, Kullu, Himachal Pradesh, India.*

ABSTRACT

The intricate process of neuroinflammation begins with the activation of glial cells and continues with the production of chemokines and cytokines that promote inflammation. Evidence suggests that neuroinflammation is prevalent in epileptic humans and animal models alike. There is a complex two-way street between neuroinflammation and drug-resistant epilepsy (DRE). On one hand, neuroinflammation helps build and sustain DRE, and on the other hand, seizures can cause neuroinflammation, which in turn causes more seizures. Many theories have been put up to clarify the connection between neuroinflammation and DRE, such as the involvement of Toll-like receptors, the NF-κB pathway, and the activation of ALK-5. Several anti-inflammatory drugs have shown benefit in animal models of epilepsy, suggesting that targeting neuroinflammation might be a potential strategy for the treatment of DRE. Nevertheless, more studies are required to ascertain the effectiveness and safety of these medicines when administered to human patients suffering from DRE. Novel treatments for this illness may be developed by understanding the processes behind the bidirectional link between neuroinflammation and DRE.

Keywords: Drug-Resistant Epilepsy, Blood-Brain Barrier, High Mobility Group Box, Toll-Like Receptor, Interleukin.

I. INTRODUCTION

A neurological illness known as epilepsy is marked by repeated seizures that do not have a known cause. [Fisher et al., 2014, Kwan et al., 2000]. Consciousness loss, changed feelings or emotions, and convulsions are among the symptoms that may result from aberrant electrical activity in the brain, which is the cause of seizures. (Kwan et al., 2000). One of the most prevalent neurological illnesses, epilepsy impacts almost 50 million individuals globally. [Lhatoo et al., 2001]. Epilepsy has many different and complicated causes. Brain traumas, infections, and other medical issues may contribute to certain instances, while heredity plays a role in others. However, the exact reason for epilepsy remains a mystery in a lot of instances. [French et al., 2004; Plioply et al., 2004]. Electroencephalography (EEG) and brain imaging scans are among the diagnostic tools used to confirm a case of epilepsy, in addition to a comprehensive medical history and physical examination. In a 2010 study, Patel et al. Medication, surgery, and nutritional therapy like the ketogenic diet are all potential treatments for epilepsy. [Palmer et al., 2004; Engel et al., 2011]. There are effective therapies for epilepsy, but many individuals still have seizures and other symptoms, which may greatly affect their quality of life. Further, those with epilepsy may have trouble getting the help they need because of the persistent stigma that surrounds the disorder. In a 2004 study, Lipoplys et al. Researchers are always looking for new ways to cure and understand epilepsy, with the hope of creating better treatments for those who suffer from it. Use of stem cells for brain tissue regeneration, creation of novel drugs for certain kinds of seizures, and non-invasive brain stimulation for seizure reduction are all areas of study that show promise. According to Scharfman and colleagues in 2007. Neurodegeneration, neuroinflammation, and blood-brain barrier dysfunction are further factors. A mystery surrounds the process of drug-resistant epilepsy. Seizures that occur repeatedly and without apparent explanation, brought on by an overabundance of electrical activity in the brain, are the hallmark of epilepsy. The epicenter of the aberrant electrical activity in the brain determines the location of the seizures. Epilepsy is significantly more common in males than in women. Banerjee et al., 2009.

II. PREVALENCE OF EPILEPSY

Whereas the incidence rate is the number of new cases in a population during a certain time period, the prevalence is the proportion of a population that is afflicted by epilepsy at any given moment. There have been many different kinds of epidemiologic research carried out in India. Included in this category are community-based door-to-door research, surveys pertaining to mental health, and studies conducted at several hospitals. Sridharan et al. (1999) The prevalence of epilepsy is estimated differently in nations with different economies. Research conducted in both industrialized and developing nations has shown that the prevalence of epilepsy is greater in the former, with over 100 cases per 100,000

people, compared to less than 100 cases per 100,000 people in the latter [Bharucha et al., 2011]. Over 10 million individuals in India are believed to be living with epilepsy (PWE). In 1999, Sridharan et al. found that it impacts around 1% of the population. People from lower socioeconomic backgrounds did not necessarily report this occurrence.

2.1 DRUG-RESISTANT EPILEPSY:

In order to obtain persistent seizure freedom, medication-resistant epilepsy must be shown by appropriate trials of two anti-epileptic drug regimens that are both tolerated and employed [Potschka H et al., 2003]. A specific definition of drug resistant, which is also used interchangeably with refractory, pharmacoresistant, and interactable epilepsy, remains unknown. It is possible to describe it as the inability to control seizures using two or three appropriate anti-seizure medications for a certain kind of epilepsy. There are a lot of theories around the pharmacological mechanism of resistance as well as the intrinsic features (lesion, first response to antiepileptic medications, and the quantity and kind of seizures prior to diagnosis). Neurobiochemical changes, cognitive deterioration, and psychosocial dysfunction are all parts of drug-resistant epilepsy (Kwan P et al., 2010). The "transporter theory," the "target hypothesis," the "network hypothesis," and the "intrinsic severity hypotheses" are the four main theories that explain pharmacoresistant. It is believed that the drug-resistant process is complex and has many unknown components, including incorrect diagnosis, medications, dosage, lifestyle choices, etc. Refractory or drug-resistant epilepsy is characterized by the ineffectiveness of conventional seizure control drugs. Up to 30% of epileptics have seizures that are resistant to drugs. In cases of drug-resistant epilepsy, seizures might persist despite taking several drugs, or they may be very difficult to manage. The inability to work, drive, or take part in everyday activities may have a major influence on a person's quality of life. A variety of variables, such as the kind of epilepsy, the intensity of seizures, and preexisting diseases, may lead to drug-resistant epilepsy. Seizure control surgery is a possibility to consider in some instances. Medical devices like a vagus nerve stimulator or responsive neurostimulation system, as well as nutritional therapy like the ketogenic diet, provide further options for treating drug-resistant epilepsy. Patients with drug-resistant epilepsy should collaborate closely with their medical providers to determine the best course of therapy.

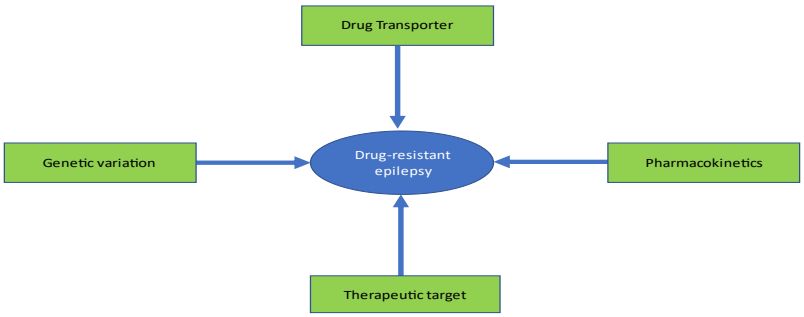

Figure-1 Drug-Resistant Epilepsy

III. NEUROINFLAMMATION

A condition known as neuroinflammation occurs when neurons, glia, and microvasculature are stimulated by the brain's natural immune response. The incidence of seizures is often associated with neuroinflammation. Neuroinflammation is a key neuropathological process in epileptogenesis, as shown by several clinical and preclinical investigations [Dey et al., 2016]. Synthesis of chemokines, ROS, cytokines, and secondary messengers promotes neuroinflammation. One possible source for these mediators or stimulators is the central nervous system, but they may also enter the brain from the bloodstream when the blood-brain barrier is broken. Neuroinflammation is a hallmark of many neurological disorders, including stroke, Parkinson's, Alzheimer's, and epilepsy [Amor et al., 2010]. The purpose of this study was to examine the existing theories on medication resistance in epilepsy and the mechanisms of drug resistance, with a focus on neuroinflammation. According to Vezzani et al. (2019), neuroinflammatory factors like IL-1β, HMGB1, transforming growth factor-β, prostaglandins, and tumor necrosis factor activate transcriptional and post-transcriptional mechanisms in brain cells, influencing neuronal, glial, and blood-brain barrier activity and functions. Traumatic brain injury (TBI), infections, age, air pollution, autoimmune diseases, spinal cord injuries, passive smoking, and other similar conditions may all trigger neuroinflammation. Environmental, genetic, disease-related, and drug-related variables all have a role in the development of drug-resistant epilepsy; neuroinflammation is one of these causes. Different diseases, traumas, infections, stresses, and toxic chemical exposures cause neuroinflammation in different ways. The binding of HMGB1 to toll-like receptor 4 (TLR4) or receptor for the advanced glycation end product (RAGE) activates proinflammatory signaling molecules, disrupts the blood-brain barrier (BBB), and upregulates the severity of seizures. This was revealed in a 2002 study by Scaffidi, P., T. Misteli, et al. [Scaffidi P et al., 2010]. Both drug-resistant temporal lobe patients and individuals with status epilepticus experience oxidative stress in their brains. Along with

this, HMGB1 is transported into neurons' and glia's cytoplasm [Ravizza T et al., 2018; Zhang S et al., 2022]. Inflammation of the nervous system is a hallmark of many neurological diseases. Neuroinflammation and neurological disorders in relation to epilepsy have been the subject of a great deal of research. Without a doubt, epilepsy is brought on by certain inflammatory disorders. One of the key components of epilepsy is the dysregulation of the inflammatory response in damaged neuronal tissue; nevertheless, the exact mechanism by which this dysregulation causes epilepsy remains unknown. Many different brain disorders may cause or exacerbate epilepsy. Numerous studies have shown that neuroinflammation is an important consequence of epilepsy. According to Lyman et al. (2014), neurons and other brain resident cells may undergo apoptosis when neuroinflammation triggers the production of several cytotoxic chemicals, including nitric oxide and reactive oxidative species. These compounds can then trigger the release of pro-apoptotic molecules. Another component that adds to neuroinflammation is the amount of iron in glial cells. Iron ferritin levels in microglia rise with age, leading to a proinflammatory dystrophic phenotype that plays a role in the development of neurodegenerative disorders. [Kempuraj et al]

When inflammation happens in the brain and spinal cord, it's called neuroinflammation. Many neurological illnesses, including epilepsy, are thought to be influenced by it. Because neuroinflammation both increases the frequency of seizures and decreases the efficacy of anti-seizure drugs, it has been shown that it may play a role in the progression of drug-resistant epilepsy. Also, neurons may be damaged or even killed off by persistent inflammation, which makes the situation much worse. Patients with drug-resistant epilepsy had higher levels of inflammatory markers as compared to individuals whose seizures were under control. One possible strategy to treat drug-resistant epilepsy and find new viable medicines is to reduce inflammation.

The significance of treating drug-resistant epilepsy with a holistic strategy that targets the inflammatory and neurological components of the disorder is underscored by the correlation between neuroinflammation and the disease.

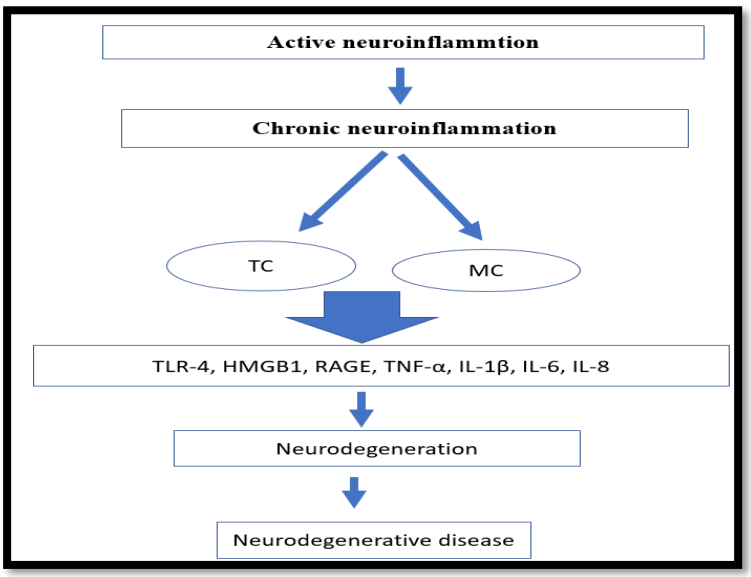

Figure-2: This is a schematic representation of neurodegeneration in the brain caused by neuroinflammation. Neuroinflammation is triggered by microglia, astrocytes, and neurons in the brain as a result of brain damage or systemic or local infections. The brain's immunological and inflammatory cells, including T-cells and mast cells, are likewise stimulated by these substances. There is an increase in the expression of inflammation-related receptor proteins in brain cells and the release of multiple proinflammatory and neurotoxic mediators when glial cells and inflammatory cells are activated. Neurodegeneration and inflammation are worsened by these inflammatory mediators and elevated protein expression, creating a self-perpetuating loop that eventually causes the illness to advance. High mobility group box 1, T-cells, TLR-4, receptor advance glycation end products, and RAGE TNF-α, also known as tumor necrosis factor-α, IL-1, IL-6, and IL-8, all of which are interleukin numbers.

3.1 Bidirectional interaction relationship between neuroinflammation and drug-resistant epilepsy:

This review study aims to delve into the connection between neuroinflammation and drug-resistant epilepsy. There is a two-way street between neuroinflammation and epilepsy; the latter may cause the former. Little is known about the two-way process by which neuroinflammation and epilepsy interact with one another. Neuromodulators, neurotransmitters, pathogen-associated molecular patterns (PAMPs), damage-associated molecular patterns (DAMPs), and their

interactions with environmental cues [Pracucci E et al., 2021]. In neuroinflammation, astrocytes and microglia are overexpressed, leading to the production of cytokines that promote inflammation, such as tumor necrosis factor (TNF)-α, interleukin (IL)-6, and IL-1β, each of which has a distinct effect on its intended recipient. Many different things may cause seizures, and seizures themselves can amplify the neuroinflammation pathway. This vicious loop causes cell death, neuronal hyperexcitability, glutamate excitotoxicity, a decreased seizure threshold, and the breakdown of the blood-brain barrier (BBB) [Sharma AA et al., 2020]. Hyperexcitability and network plasticity are both impacted by neuroinflammation, which has been highlighted in the review (Vezzani et al., 2019). This review will evaluate the current state of research on neuroinflammation as a biomarker for the onset of drug-resistant epilepsy and the efficacy of treatment interventions that aim to target the receptors and starting elements of these pathways. By focusing on these receptors and starting factors, we may change illness, prevent epilepsy, and ictogenesis. Potentially unexplored therapeutic targets in the fight against epilepsy include neuroinflammation.

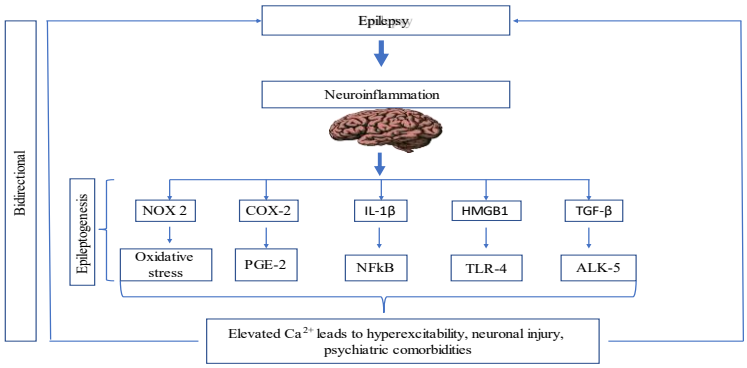

Figure-3: It is shown that epilepsy causes inflammation in the nervous system, which activates many proteins including NOX2, COX2, IL-1β, HMGB1, and TGF-β. Additionally, it induces oxidative stress, PGE2, NFκB, TLR-4, and ALK-5. A vicious cycle of hyperexcitability, neuronal damage, and mental health issues results from this high calcium level, which in turn produces epilepsy.

IV. ROLE OF HMGB1

A structural protein in the nucleus of eukaryotic organisms, the high mobility group (HMG) protein bends, changes, and modifies the structure of DNA. A large portion of the HMG superfamily consists of members from the HMGA, HMGB, and HMGN families. Three proteins, HMGB1, HMGB2, and HMGB3, comprise the HMGB family (formerly called HMG4 or HMG2B) [Dai et al., 2021]. Proteins that are not histones but are essential for the structure of DNA, known as high mobility group box 1 proteins, are very conserved and belong to the class of DAMPs. HMGB1 responds to epileptogenic stresses by acting as an inflammatory cytokine. Under neuroinflammatory circumstances, inflammasomes are activated by neurons and glial cells, leading to the active production of HMGB1. This, in turn, activates at least two surface receptors on target cells, namely TLR4 and RAGE. Ravizza et al., 2018. HMGB1 has the ability to bind to a variety of receptors, including Toll-like receptors 2, 4, and the receptor for advanced glycation end products (RAGE). HMGB1 boosts inflammation by activating pathways that encourage chemotaxis and cytokine production, such as nuclear factor-κB (NF-κB). Yang et al. (2017), HMGB1 is thought to have an impact on the development of a number of diseases. These include encephalitis, TBI, cerebral ischemia, sepsis, arthritis, and respiratory problems. It is possible for certain damaging signals to trigger nuclear production of HMGB-1, which may then be transferred to the cytoplasm. A number of chemicals, including late glycation end products (RAGE), Toll-like receptor 2, and Toll-like receptor 4, are generated when it interacts with these receptors (TLR4). To control inflammation caused by HMGB1, HMGB1 secretion must be regulated. Several mechanisms are required for this regulation, including methylation, acetylation, and calcium-dependent protein kinase C phosphorylation. Recent research found that HMGB1 and TLR4 were implicated in the first and subsequent occurrence of seizures in rats that were tested [choi et al., 2011]. The study found that glia and neurons in epileptogenic tissue had higher levels of IL-1β, HMGB1, and its receptors IL-1R1 and TLR4 after analyzing brain samples from individuals with drug-resistant epilepsy. Human epilepsy may include many signaling pathways [Vezzani et al., 2014]. As an endogenous TLR4 stimulator, high mobility group box-1 is an important player (HMGB1). Following TLR4 activation, the downstream pathway is mostly activated via a route that depends on MyD88. The activation of a TLR4 attracts MyD88, which in turn leads to the ubiquitination of TRAF6 and the activation of the NF-B inflammatory pathway. Translation of the pro-inflammatory cytokines IL-1β and TNF-α into the nucleus occurs due to the activation of NF-κB [Zhang et al., 2018]. It is possible for necrotic cells and living cells including macrophages, neurons, astrocytes, and hepatocytes to passively release HMGB1 in response to tissue injury. When there is ischemia or brain trauma, the neuronal nucleus releases HMGB1, which compromises the BBB and increases inflammatory reactions in the brain. Pericytes and vascular endothelial cells are also stimulated to contract

when it is present. More and more evidence is suggesting that HMGB1 translocation from the brain into peripheral circulation is a key mediator of the inflammatory response in epileptogenesis [Fu et al., 2017]. Its association with epilepsy is the reason for the current surge of interest in HMGB1. The exact method by which HMGB1 contributes to epileptogenesis remains unclear, but it is believed to do so mostly by triggering inflammatory processes and rupturing the BBB. The role of HMGB1 in the development of epilepsy has been the subject of a great deal of prior research. [**Paudel et al., 2018**].

V. Neuroinflammation and dysfunction of the Blood-Brain Barrier (BBB)

The BBB is an intricate and ever-changing barrier that separates the central nervous system from the bloodstream. It is composed of brain endothelial cells, pericytes, and astrocytic endfeet, which come together to create a neurovascular unit (NVU). The NUV is encased in the basal lamina and is structured by several cell types. Abbott et al. (2010), Hawkins et al. (2005), and Abbott et al. (2013) all agree that ECM proteins are crucial for their development and maintenance. Macromolecular large molecules such as proteins and lipids are mostly excluded by intercellular TJs (occludins, claudins, and junctional adhesion molecules). [Liebner S et al., 2018] The brain is able to keep small compounds that are insoluble in lipids out of the brain. The BBB, also known as the multidrug resistance (MDR) gene, encodes a number of proteins that play a role in the control of the absorption of drugs into the brain and the elimination of harmful lipophilic metabolites that enter the bloodstream [Lazarowski et al., 1999, Sisodiya et al., 2001, Sisodiya et al., 2002]. Endothelial cells, astrocytes, and pericytes are all part of the neurovascular unit. In order to promote inflammatory responses and activate Toll-like receptor 4 (TLR4) and receptor for Advance Glycation End Product (RAGE), neurons, glia, and microglia produce damage-associated molecular protein (DAMP,s). Afterwards, pro-inflammatory cytokines including IL-1β and NFκB, which are nuclear factor kappa light chain enhancers of activated B cells, show an increase in expression. Seizures and neuroinflammation are both accelerated by these proinflammatory cytokines. The activation of these signals causes the blood-brain barrier (BBB) to malfunction, which in turn increases the expression of the ABCB1 gene, which in turn causes an increase in P-gp expression and, ultimately, drug-resistant epilepsy [Bedolla et al., 2022]. In a similar vein, a mountain of data points to neuroinflammation as a key player in BBB disruption and dysfunction, which in turn sets off a cascade of events leading to DRE. It has been suggested by [Löscher et al. 2020] that inflammatory mediators might potentially contribute to three things may lead to seizures that are resistant to drugs: (i) oxidative stress, (ii) aberrant angiogenesis leading to "leaky" arteries, and (iii) activation of transcytosis or failure of the blood-brain barrier. [**Loscher et al., 2020, sisodiya et al., 2002, Loscher et al., 2005**].

Drug-resistant seizures may benefit from inflammatory mediators in three ways. Astrocytes may emit inflammatory compounds due to BBB permeability changes. Astrocytes' inflammatory state is crucial to these processes. This self-perpetuating cycle causes seizures, cell death, and neural network stiffness, increasing the disease's "intrinsic severity". When the BBB fails, albumin may extravasate into the brain parenchyma, which may increase the "buffering" effect of albumin binding to pharmaceuticals and minimize the amount of unbound drugs in functionally relevant brain areas. P-gp activation in endothelial cells and maybe perivascular astrocytes via COX2-PGE2-EPIR and IL-1beta, IL-1R1 inflammatory pathways supports the transporter hypothesis of drug resistance. Third, post-translational alterations generated by inflammatory mediators may impair voltage-gated and receptor-operated ion channel sensitivity to ASD targets, supporting the pharmacodynamic (target) notion of medication resistance. [**Löscher et al., 2020**.]

5.1 Role of Toll-Like Receptors (TLR):

One group of proteins called toll-like receptors (TLRs) are responsible for identifying certain chemical patterns, such as those linked with pathogens or damage. Kwai et al. (2007). Membrane receptors are present on many different types of cells, including immune cells like dendritic cells and macrophages. A signaling cascade that activates immune cells and produces inflammatory cytokines is set in motion when a TLR identifies a PAMP or DAMP [Akashi et al., 2008]. As a line of protection against pathogens and damaged tissues, this reaction is crucial. Among humans, there are ten distinct toll-like receptors (TLRs), and each one is responsible for identifying a unique set of infections and triggering an appropriate immune response.

1. TLR1: Recognizes bacterial lipoproteins and forms heterodimers with TLR2. [Takeuchi et al., 2009].
2. TLR2: Recognizes bacterial lipoproteins, peptidoglycans, and lipoteichoic acids. Forms heterodimers with TLR1 and TLR6. [Takeuchi et al., 2009].
3. TLR3: Recognizes viral double-stranded RNA. [Alexopoulou et al., 2001]
4. TLR4: Recognizes bacterial lipopolysaccharides (LPS) and certain viral proteins. [Beutler et al., 2000]
5. TLR5: Recognizes bacterial flagellin. [Hayashi et al., 2001]
6. TLR6: Recognizes bacterial lipoproteins and forms heterodimers with TLR2. [Takeuchi et al., 2009]
7. TLR7: Recognizes viral single-stranded RNA. [Heil et al., 2004].
8. TLR8: Recognizes viral single-stranded RNA and certain synthetic compounds. [Heil et al., 2004]
9. TLR9: Recognizes unmethylated CpG DNA motifs in bacterial and viral DNA. [Bauer et al., 2001]
10. TLR10: The ligand specificity of TLR10 is not yet fully understood. It has been suggested to form heterodimers with TLR1 or TLR2 and modulate their signaling. [Chuang et al., 2004].

Proteins known as toll-like receptors (TLRs) are essential for the immune system to react to harmful invaders. It aids in the clearance of infections by recognizing and binding to certain molecules on the surface of microbes such viruses, bacteria, and others [Vezzani et al.,2015]. Recent studies, however, have linked TLRs to neuroinflammation and drug-resistant epilepsy [Maroso et al., 2010].

Seizures caused by drug-resistant epilepsy cannot be managed with medication. Up to 30% of epileptics experience it, and it may greatly diminish their standard of living. Although the precise reason for drug-resistant epilepsy remains a mystery, it is believed to be associated with alterations in the brain's immune system and inflammation. Citation: Liberzzi et al., 2012. Neurons and glial cells are among the many brain cells known to express TLRs, suggesting that they may be involved in these functions. [Maroso et al., 2010].

A variety of brain immune cells, such as microglia and astrocytes, get activated during neuroinflammation, a complicated process [Vezzani et al., 2011]. Many neurological disorders, such as epilepsy, Alzheimer's disease, and multiple sclerosis, are believed to be influenced by it. Neuroinflammation may have its roots in toll-like receptors (TLRs), which are thought to have a role in the activation of certain immune cells [Hanke et al., 2013].

New research points to TLR targeting as a potential strategy for treating neuroinflammation and drug-resistant epilepsy. Some studies have shown that activating TLR2 has anti-inflammatory effects in the brain, whereas others have demonstrated that inhibiting TLR4 reduces seizures in animal models of epilepsy [Vezzani et al., 2011, Hanke et al., 2013]. To create safe and effective treatments that target TLRs, more study is required to understand their function in these diseases.

Potentially useful for disease-modifying epilepsy treatment is targeting the TLR4 signaling system. It is important to consider TLR4 receptor ligands when developing strategies to reduce excitability and seizure susceptibility increases mediated by TLR4. It is intriguing that in this context, the inducible heat shock protein 70 (HSP70) was shown to be up-regulated in the hippocampus and para-hippocampal cortex in a rat post-status epileptics model with epilepsy manifestation following a latency period [Walker et al., 2016]. The role of HSP70 in modulating TLR4 activity was previously shown [Vabulas et al., 2002]. It was recently shown to be useful in a kindling model; mice that overexpressed human HSP70 had heightened susceptibility to seizures, lower thresholds, and early onset of generalized seizures when stimulated.

5.2 Role of NFκB:

A transcription factor known as nuclear factor kappa B (NF-κB) is essential for controlling inflammatory reactions [Kaltschmidt et al., 2009]. It has a role in the synthesis of adhesion molecules, pro-inflammatory cytokines, and chemokines, and in the activation of immune cells. According to Vezzani et al. (2011), NF-κB has a role in controlling cell survival as well as apoptosis.

It has been shown that NF-κB is activated in both animal models of epilepsy and human patients with the disorder. According to Falcao et al. (2014) and Chen et al. (2014), it has a role in controlling inflammation caused by seizures and the resulting harm to the blood-brain barrier. The activation of NF-κB has also been associated with the overexpression of the inflammasome and Toll-like receptors (TLRs), two important components of the innate immune response [Vezzani et al., 2013]. Animal models of epilepsy have shown that NF-κB inhibition decreases inflammation and seizure activity. It follows that focusing on NF-κB signaling pathways might be a promising approach to treating epilepsy and related conditions.

When glial cells like microglia and astrocytes become activated and release pro-inflammatory cytokines and chemokines, a disease known as neuroinflammation develops in the CNS [Vezzani et al.,2019, Falcao et al., 2014]. Several neurological diseases, including drug-resistant epilepsy (DRE), include this pathway in their pathophysiology. The transcription factor NFκB is vital in controlling immunological responses and inflammation (Iyer et al., 2012). According to Vabulas et al. (2002), it controls the expression of genes related to inflammation, cell survival, and proliferation when triggered by oxidative stress, pathogens, and pro-inflammatory cytokines. Neuroinflammation and DRE are both impacted by NFκB. Multiple investigations have shown that epileptic patients, especially those with DRE, have elevated NFκB activity in their brains. As an example, a research conducted by Lorigados et al. (2010) indicated that NFκB activation was higher in the hippocampus of individuals diagnosed with temporal lobe epilepsy (TLE) when compared with controls. According to Wang et al. (2017), NFκB activation was discovered to be elevated in the brains of rats suffering from pilocarpine-induced epilepsy, a kind of transient ischemic epilepsy. Drug resistance in epilepsy has also been linked to NFκB. Brain tissue from individuals with drug-resistant epilepsy showed lower levels of NFκB activation compared to those from drug-responsive epilepsy, according to one research. In a mouse model of DRE, this research also discovered that controlling seizures was enhanced by blocking NFκB activation [Kaltschmidt et al., 1997]. The pathophysiology of several neurological illnesses, such as Alzheimer's disease and Parkinson's disease, has been linked to NFκB, along with its involvement in neuroinflammation and DRE (Ghosh et al., 2007). Taken together, our results point to NFkB as a key player in the development of neuroinflammation and DRE. As a potential treatment option for several diseases, targeting NFkB has great promise.

5.3 Role of IL-1β:

An important regulator of inflammation and immunological responses is the pro-inflammatory cytokine IL-1beta. According to Zhang et al. (2022), it has a role in the development of neuroinflammation and a number of neurological illnesses, including epilepsy. It is generated by activated glial cells, such as microglia and astrocytes.

According to Yamanaka et al. (2022), there have been several studies that have shown elevated levels of IL-1beta in the brains of epileptic patients, especially those with drug-resistant epilepsy (DRE). In contrast to healthy controls, individuals suffering from temporal lobe epilepsy (TLE) had elevated levels of interleukin-1beta in their hippocampus, according to one research [Vezzani et al., 2000]. A different research indicated that rats suffering from pilocarpine-induced epilepsy, a kind of TLE, had elevated levels of IL-1beta in their brains. Additionally, IL-1beta has been linked to the development of epilepsy medication resistance [Ravizza et al., 2007]. Research in mice using a DRE model indicated that blocking IL-1beta signaling helped with seizure management (Vezzani et al., 2012). Shaftel et al. (2008) and Mogi et al. (1994) found that IL-1beta is involved in neuroinflammation and epilepsy, but it has also been linked to the development of other neurological illnesses, such as Alzheimer's and Parkinson's. We conclude that IL-1beta is an important player in the development of neuroinflammation and epilepsy based on these results. A possible effective treatment approach for these diseases might be to target IL-1beta.

5.4 Role of ALK-5 and TGF-β:

The protein ALK-5 is a member of the receptor family for transforming growth factor-beta (TGF-β). Cell migration, differentiation, and proliferation are just a few of the many cellular activities in which it participates. According to research, the ALK-5 pathway might be involved in the onset of epilepsy and other neurological diseases including Alzheimer's and Parkinson's [Von et al., 2015]. As a result, epilepsy and other neurological illnesses may be amenable to therapy via focusing on the ALK-5 pathway.

5.5 Role of COX-2 and PGE-2:

Inflammatory responses are mediated by prostaglandins, which are produced when arachidonic acid is converted to them by an enzyme in the metabolic pathway known as cyclooxygenase (COX). According to Zhu et al. (2020), inducible cyclooxygenase-2 (COX-2) activation is believed to be a factor that triggers neuroinflammation in the brain. According to Oliveira et al. (2008), three COX isozymes were discovered and named COX-1, COX-2, and COX-3.

According to Rumia et al. (2012), PGE2 is the most abundant prostaglandin generated centrally and peripherally in models of both acute and chronic inflammation.

Seizure and epilepsy development-related cyclooxygenase-2 (COX-2) participation has garnered a lot of research because of its role as a major connection to several inflammatory processes. According to Zhu et al. (2020), after inducing seizures, COX-2 is shown to be elevated in several brain cells. This leads to an increase in the synthesis of proinflammatory mediators, or PGs, which in turn makes the seizures worse.

It is well-established that COX-2 and PGE2 contribute to epilepsy. A number of research have looked at how this chemical contributes to epilepsy and how it develops over time.

In a study conducted by Dannhardt et al. in 2001, it was shown that rats with induced epilepsy had elevated COX-2 expression in their hippocampus. The researchers also observed that seizures were less severe when the rats were treated with a COX-2 inhibitor. Treatment with a PGE2 receptor antagonist decreased seizure frequency in individuals with epilepsy, according to another study. The researchers found that PGE2 levels were elevated in the cerebral fluid of these patients. Reference: Matsuoka et al., 2005. Furthermore, a research discovered that PGE2 and COX-2 contribute to the development of TLE, a prevalent kind of epilepsy. The research found that COX-2 and PGE2 levels were elevated in the hippocampus of TLE patients. Additionally, it was shown that animal models of TLE had seizures less often and for shorter durations after being treated with a COX-2 inhibitor [Chen et al., 2018]. The results of this study point to COX-2 and PGE2 as possible epilepsy therapy targets. To create effective treatments that target these molecules, however, more study into their roles in epilepsy is required.

VI. RELATIONSHIP BETWEEN OXIDATIVE STRESS AND NEUROINFLAMMATION

Neuron degeneration, a hallmark of many brain diseases, is caused by oxidative stress damage, which includes free radicals, glycated products, and lipid peroxidation. Source: Popa et al. (2013). The production of reactive oxygen species (ROS) in brain tissues has the potential to impact neuronal transmission both synaptic and nonsynaptic, leading to neuroinflammation, cell death, neurodegeneration, and memory loss. The correlation between inflammation and oxidative stress has been extensively studied by several writers. There is mounting evidence that chronic inflammatory disorders are harmful due to oxidative stress. In the inflammatory response, IL-2 and activated T lymphocytes draw in neutrophils and macrophages, which then release a flood of reactive oxygen species (ROS) that kill the bacteria they ingest (oxidative burst) [Lu SP et al., 2007]. Microglia and astrocytes in the brain create reactive oxygen species (ROS), which affect neuronal and glial nonsynaptic communication. An unbalanced immunological response, inflammation, and susceptibility to infection are caused by reduced levels of tripeptide glutathione (GSH), an intracellular thiol antioxidant. In 2011, Ghezzi et al. had published their findings. Instead of thinking about inflammation as an illness,

one should see it as a natural biological activity. Neurodegenerative disorders like epilepsy are caused by a decrease in hippocampus neurogenesis, which in turn is caused by an inflammatory response and oxidative stress. Consequently, these processes need more research for the therapeutic treatment of these diseases. Research on both humans and animals has shown that epileptics are more susceptible to inflammation. For instance, regions of the brain responsible for the start and progression of seizures have a marked inflammatory response when subjected to experimentally produced seizures. [Vezzani et al.,2005].

6.1 Neuroinflammation as a target for drug-resistant epilepsy:

The efficacy of treating neuroinflammation in drug-resistant epilepsy has been mostly unproven. To further explore neuroinflammation as a potential target for drug-resistant epilepsy, further study is required. Seizure reduction has been achieved with the use of several neuroinflammation-targeting medications and agents, such as IL-1β inhibitors, COX-2 inhibitors, and EP1 and EP2 receptors inhibitors (see TABLE 1). The inflammatory response in epilepsy is multifaceted and may vary from one patient to another. According to Garcia C et al. (2021-22), these variations may provide light on why drug-resistant epilepsy is so burdensome and highlight the need of developing customized therapies. To treat drug-resistant epilepsy, neuroinflammation may be an excellent target. In this review, we will go over the basics of drug-resistant epilepsy, including what it is, how it's caused, and how to treat neuroinflammation. A number of potential targets for the treatment of drug-resistant epilepsy exist along the pathway of neuroinflammation, including HMGB1, TLR-4, RAGE, IL-1R, COX-2, IL-6, Chemokine receptor, NLRP, NMDA, and NR2B subunit, among others. Additionally, there may be a vicious cycle of neuroinflammation that involves both epilepsy and neuroinflammation. Although there are a few anti-inflammatory medications that work by conventional methods, like aspirin, that are used to treat epilepsy, the newer generation of anti-inflammatory and anti-epileptic medications has made their moderate benefits more noticeable. [Parsons et al., 2022].

TABLE 1. Overview of studies targeting neuroinflammation to treat drug-resistant epilepsy.

Drugs	Mechanism of drug/ target	Outcome
Anakinra	IL-1 Receptor antagonist	Reduced the number of seizures, Reduced peripheral blood monocyte cytokine production such as IL-1 IL-10 [Singh et al., 2014]
Aspirin	COX-2 inhibitor	Reduced seizures frequency and duration Reduced proinflammatory cytokines These studies lack inflammation analysis [Bialer M et al., 2020]
Adalimumab	Anti-TNF monoclonal antibod	Reduced TNF-α levels [Lagarde et al.,2016]
Tocilizumab	IL-6 Receptor	IL-6 levels normalized Patients experienced severe adverse events related to infection during epilepsy [Jyonouchi H et al., 2016]
Anakinra+ canakinumab	IL-1R antagonist, Monoclonal antibody against IL-1 Receptor	Reduced IL-1-driven systemic autoinflammation [Kenney et al., 2016]
Minocycline	Microglial activation inhibitor	Inhibit microglial activation Reduced hippocampal neuroinflammation Reduced seizures frequency, severity, and duration These studies lack inflammation analysis [Godfred R.M. et al., 2013

6.2 Oxidative stress as a target for drug-resistant epilepsy:

When used alone or in conjunction with current AEDs, antioxidant treatments for neurological diseases like epilepsy show encouraging results. Additionally, there are a number of reasons why antioxidants have not been used therapeutically to improve neuroprotection in epilepsy. For example, many treatments only target one type of oxidative stress, and other antioxidants are site-specific. The increasing interest in treating epilepsy means that novel compounds with enhanced pharmacological characteristics are continually being investigated in the clinic, even though this is a relatively young subject. Table 2 below lists the antioxidant molecules; Gluck et al. (2000) found that GSH is an antioxidant molecule in the cortex, whereas SOD is necessary for neuroprotection against OS in the hippocampus.

TABLE 2. Targeting oxidative stress to treat drug-resistant epilepsy

ANTIOXIDANT MOLECULES	MECHANISM OF ACTION	EFFECTS	REFERENCES
Coenzyme Q10	Increases TCA and antioxidant enzyme levels	SOD and GSH levels increased, and lipid peroxidation reduced.	[Shin et al., 2005]
Vitamin E	Peroxyl radical scavenger	Increased levels of glutathione and catalase and enhanced antioxidant capacity.	[Mehvari et al., 2016]
Cannabidiol	GRP55 inhibitor inhibits adenosine uptake	Reduced ROS production; Enhanced antioxidant defenses	[Arzimanoglou et al., 2020]
Naringenin	Free radical scavenger	Increased levels of glutathione and antioxidant enzymes	[Shakeel et al., 2017]
N-acetylcysteine	Glutathione precursor reduced	Glutathione homeostasis impairment reduced.	[Yang et al., 2020]
Sulforaphane	Stimulate NRF2 pathway	Malondialdehyde levels reduced while glutathione levels increased.	Sandouka et al., 2021]
Curcumin	Metal chelator and Free radical scavenger	The enzyme superoxide dismutase is now more active. The expression of inflammatory cytokines and chemokines were decreased.	[Dhir et al., 2018]

VII. CONCLUSION:

We found in this review that oxidative stress and neuroinflammation may play a major role in epilepsy and contribute to treatment resistance. According to the two-way interaction processes, neuroinflammation has the potential to generate excitotoxicity, which in turn causes epilepsy by upregulating proinflammatory cytokines. However, a large body of research indicates that neuroinflammation is a direct result of epilepsy, suggesting that the two processes may be mutually reinforcing. Epilepsy may develop in the central nervous system (CNS) via a number of different neuroinflammatory pathways, including those involving HMGB1/TLR4 and TRPM2 signaling pathways, genetic alterations (PCDH19 and mTOR), activation of the maternal immune system, brain injury ischemia, and so on.

There is a connection between neuroinflammation and the onset of epilepsy. One major inflammatory component is high mobility group protein B1 (HMGB1), which is known to have elevated expression and a larger fraction of translocation from the nucleus to the cytoplasm.

Neuroinflammation may be triggered by oxidative stress. New evidence has connected inflammation to a process involving the generation of reactive species and the subsequent redox alterations.
In addition, the fact that HMGB1 might be a therapeutic target provides insight into how HMGB1 signaling contributes to drug-resistant epilepsy.

In response to HMGB1 production by astrocytes, microglia activate IL-1, a proinflammatory cytokine, via Toll-like receptors 4 and RAGE. This, in turn, may cause NMDA receptor NR2B subunit phosphorylation, which in turn causes glutamate release, hyperexcitability, and ca2+ influx. Possible elevation of HMGB1 expression in hyperexcitability.

- Here, hyperexcitability triggers HMGB1 upregulation, which in turn triggers hyperexcitability, creating a vicious cycle.
- Research has connected oxidative stress to the activation of neuroinflammation. But neuroinflammation increases oxidative stress. Similarly, this route repeats itself in an endless loop.

Neurons and glia in status epilepticus patients undergo cytoplasmic translocation of HMGB1, a process linked to oxidative stress in the brain. Research has connected oxidative stress to the activation of neuroinflammation. Conversely, neuroinflammation may trigger the secretion of many cytotoxic substances, such nitric oxide and reactive oxidative species. These cytotoxic chemicals in turn can trigger the production of pro-apoptotic compounds, which can cause neurons and other brain resident cells to undergo apoptosis.

References

1. Abbott, N. J., Patabendige, A. A., Dolman, D. E., Yusof, S. R., & Begley, D. J. (2010). Structure and function of the blood–brain barrier. *Neurobiology of Disease*, *37*(1), 13–25. https://doi.org/10.1016/j.nbd.2009.07.030
2. Banerjee, P. N., Filippi, D., & Hauser, W. A. (2009). The descriptive epidemiology of epilepsy—A review. *Epilepsy Research*, *85*(1), 31–45. https://doi.org/10.1016/j.eplepsyres.2009.03.003
3. Chen, S., Zhang, X., & Yang, H. (2018). COX-2 inhibitors reduce epileptogenesis by inhibiting neuroinflammation in a rat model of temporal lobe epilepsy. *Journal of Neuroinflammation*, *15*(1), Article 225. https://doi.org/10.1186/s12974-018-1262-4
4. Fisher, R. S., Acevedo, C., Arzimanoglou, A., Bogacz, A., Cross, J. H., Elger, C. E., Engel, J., Forsgren, L., French, J. A., Glynn, M., Hesdorffer, D. C., Lee, B. I., Mathern, G. W., Moshé, S. L., Perucca, E., Scheffer, I. E., Tomson, T., Watanabe, M., & Wiebe, S. (2014). ILAE official report: A practical clinical definition of epilepsy. *Epilepsia*, *55*(4), 475–482. https://doi.org/10.1111/epi.12550
5. Kwan, P., & Brodie, M. J. (2000). Early identification of refractory epilepsy. *New England Journal of Medicine*, *342*(5), 314–319. https://doi.org/10.1056/NEJM200002033420503
6. Löscher, W., & Potschka, H. (2005). Role of drug efflux transporters in the brain for drug disposition and treatment of brain diseases. *Progress in Neurobiology*, *76*(1), 22–76. https://doi.org/10.1016/j.pneurobio.2005.04.006
7. Lyman, M., Lloyd, D. G., Ji, X., Vizcaychipi, M. P., & Ma, D. (2014). Neuroinflammation: The role and consequences. *Neuroscience Research*, *79*, 1–12. https://doi.org/10.1016/j.neures.2013.10.001
8. Maroso, M., Balosso, S., Ravizza, T., Liu, J., Aronica, E., Iyer, A. M., Rossetti, C., Molteni, M., Casalgrandi, M., Manfredi, A. A., Bianchi, M. E., & Vezzani, A. (2010). Toll-like receptor 4 and high-mobility group box-1 are involved in ictogenesis and can be targeted to reduce seizures. *Nature Medicine*, *16*(4), 413–419. https://doi.org/10.1038/nm.2127
9. Potschka, H. (2010). Transporter hypothesis of drug-resistant epilepsy: Challenges for pharmacogenetic approaches. *Pharmacogenomics*, *11*(10), 1375–1385. https://doi.org/10.2217/pgs.10.121
10. Ravizza, T., Terrone, G., Salamone, A., & Vezzani, A. (2018). High mobility group box 1 is a pathogenic factor and a mechanistic biomarker of epilepsy. *Brain, Behavior, and Immunity*, *72*, 8–17. https://doi.org/10.1016/j.bbi.2017.10.008
11. Scaffidi, P., Misteli, T., & Bianchi, M. E. (2002). Release of chromatin protein HMGB1 by necrotic cells triggers inflammation. *Nature*, *418*(6894), 191–195. https://doi.org/10.1038/nature00858
12. Sharma, A. A., & Sorkhabi, M. (2020). Neuroinflammation and blood-brain barrier dysfunction in epilepsy: A systematic review. *Epilepsy & Behavior*, *112*, Article 107356. https://doi.org/10.1016/j.yebeh.2020.107356
13. Vezzani, A., Balosso, S., & Ravizza, T. (2019). Neuroinflammatory pathways as treatment targets and biomarkers in epilepsy. *Nature Reviews Neurology*, *15*(8), 459–472. https://doi.org/10.1038/s41582-019-0217-x
14. Vezzani, A., French, J., Bartfai, T., & Baram, T. Z. (2011). The role of inflammation in epilepsy. *Nature Reviews Neurology*, *7*(1), 31–40. https://doi.org/10.1038/nrneurol.2010.178
15. Zhang, S., & Qin, C. (2022). The role of IL-1β in neuroinflammation and epilepsy: New insights into therapeutic strategies. *Frontiers in Immunology*, *13*, Article 845839. https://doi.org/10.3389/fimmu.2022.845839