Chapter

Molecular Mechanisms behind Initiation of Focal Seizure in Temporal Lobe Epilepsy: Computational Study

Ruchi Jakhmola Mani and Deepshikha Pande Katare

Abstract

Epilepsy is a noncommunicable disease of the brain that affects people of all ages. The chapter aims to identify protein targets and their mechanism of action behind temporal lobe epilepsy. Differentially expressed proteins in temporal lobe epilepsy (TLE) were used to derive a hypothesis demonstrating routes of protein interactions causing focal seizure and identification of putative target receptor for its treatment. Text mining was done by constructing a Boolean query with keywords such as temporal lobe epilepsy, focal seizures, proteomics, etc., in different scientific search engines. The proteins were further used for creating protein interaction network and analysed for their role in focal epileptic seizure pathway. The most appropriate route for initiation of seizure was observed to be route 3. It describes the dysregulated signal transduction from adenosine A1 receptor (ADORA1) to gamma-aminobutyric acid (GABA) B receptor 1 (GABBR1). This causes electrical imbalance and hyper-excitation of neurons that lead to focal seizure. The study also predicts that YWHAZ (3-monooxygenase/tryptophan 5-monooxygenase activation protein zeta) could be the potential target for preventing focal seizures. The network framed in this study is ideal for studying the cascades of events that may occur during focal seizures in TLE and is useful in drug discovery.

Keywords: temporal lobe epilepsy, focal seizures

1. What is epilepsy?

Epilepsy is the world's fourth most common neurological disorders. As indicated by the International League Against Epilepsy (ILAE), seizure is a transient of signs or appearances on the account of irregular, over the top, or synchronous neuronal development in the cerebrum. According to the WHO, 50 million people worldwide have epilepsy, and it is the fourth most common disease after Alzheimer's [1]. Epilepsy is an electrical unevenness in the cerebrum because of a few reasons like inactive way of life, hereditary distortions, hypoxic conditions, cranial harm, aggravation, or expansion in oxidative anxiety levels in the body, et cetera. A seizure is a sudden surge of electrical movement in the cerebrum. Seizures control how a man shows up or represents a brief span period. Neuronal cells either energize or repress

(stop) other neuronal cells from sending messages. As a rule, there is an equalization of cells that energize and stop these messages. However, when a seizure occurs, there might be a lot of or too little action, bringing about an imbalance in the middle of exciting and halting movement. The nature of seizures varies, because the lobes of the brain control different behaviors, movements, and experiences. The accurate reason for a seizure is still obscure.

Some common causes of epilepsy are as follows:

- Low oxygen during birth
- Head injuries that occur during birth or due to accidents during youth or adulthood
- Brain tumors
- Genetic conditions that result in brain injury, such as tuberous sclerosis
- Infections such as meningitis or encephalitis

Because epilepsy is caused by abnormal activity in brain cells, seizures can affect any process your brain coordinates. Seizure signs and symptoms may include:

- Temporary confusion
- A staring spell
- Uncontrollable jerking movements of the arms and legs
- Loss of consciousness or awareness
- Psychic symptoms

Symptoms vary depending on the type of seizure. In most cases, a person with epilepsy will tend to have the same type of seizure each time, so the symptoms will be similar from episode to episode.

2. Focal seizures

Epilepsy comprises of more than 40 clinical disorders and is characterized by repeated seizures. Approximately 30% of patients treated with antiepileptic drugs (AEDs) are lacking control on seizures [2]. Seizure is a transient state of electrical imbalance in the brain. There are different types of seizures such as generalized, focal, and other types which hinder the normal functioning of the brain and cause epilepsy. Temporal lobe epilepsy is the most common seizure disorder in adults. The main cause for TLE is hippocampal sclerosis, and it constitutes around 80% of TLE cases [3]. The incidence rate of TLE was reported as 10.4 per 100, and its prevalence is 1.7 per 1000 people [4, 5]. TLE can be sporadic, ordinarily with a positive family history, or it can give clear familial repeat [6]. There is 70% of neuronal loss within the hippocampal region along with repeated focal seizures [7]. The regular clinical signs of focal seizures include gazing, absence of responsiveness, and mouth or hand automatisms. Focal seizure is mainly caused by the malfunction of ion channels. External stimuli like infection, tumors, and cranial injuries upregulate

adenosine and increase the permeability of the blood-brain barrier which results in excessive influx of neurotransmitters into the hippocampal region. Lesser reuptake of these inhibitory neurotransmitters like gamma-aminobutyric acid (GABA) and glutamate by the receptors from the synaptic cleft causes neuroexcitation [8].

A recent study showed that GABA levels were found to be lower in diseased state than normal conditions. GABA system and receptor agonists have been used in the past for decreasing the effect of a seizure [9]. AEDs such as benzodiazepines (including clonazepam and clobazam) increase the frequency of opening of GABA receptors, and barbiturates (such as phenobarbitone) increase the time period for receptor opening. Thus, these drugs affect the release and movement of GABA [10]. Besides their positive effects on cognition ability and behavioral domain, AEDs have adverse effects on human health.

In this chapter we have hypothesized a protein-protein interaction network which proposes a possible mechanism underlying a focal seizure at a molecular level. Several proteins in this pathway were differentially regulated which initiates a cascade of events downstream that negatively regulates GABA [11]. The downregulation of GABA causes electrical imbalance in the brain and hence leads to focal seizure. Although drugs are available for treating TLE, prevention from its side effects is questionable; therefore, we have proposed new receptor for preventing seizures.

3. Building a hypothesis

3.1 Building a local protein database for network assessment

Text mining was done by constructing a Boolean query with keywords such as temporal lobe epilepsy, focal seizures, proteomics, differential expression, and human in different scientific search engines and databases like PubMed Central, ScienceDirect, and Google Scholar. Several combinations of keywords were used to fetch research articles according to the study. Many research articles were obtained and screened manually for differentially expressed proteins in TLE. The proteins were further used to create protein-interaction network.

3.2 Creation of protein interaction network

Protein accession numbers were submitted to Cytoscape v2.6 software as query, and it gave a master network between the query proteins. The master network comprised of query proteins and their interacting partners. This network was created by BioGRID, IntAct, and other protein-protein interaction databases included in the Cytoscape framework. This network was accompanied with the most followed pathways ranked according to their e-values. E-value is an indicator that the results are independent of the protein queries chosen and the master network is unbiased i.e. the network is based on experimentally verified protein-protein interactions and hence are reflected in the network.

3.3 Framing and analysis of hypothesis

The master network was studied, and the proteins exclusively responsible for a focal seizure were extracted and linked to formulate a pathway that might be responsible for causing seizure in an epileptic brain. This hypothesis was formulated in a stepwise manner, and it explains the series of interactions that might occur in the brain during the initiation of a focal seizure in TLE.

4. Results

Proteins involved in TLE were collected after extensive literature survey. The network is in the form of ball and sticks. Balls represent protein nodes, and sticks represent the interaction between them. The proteins immediately next to the query proteins are called as first neighbors. These first neighbors are later connected to second neighbors and so on (**Figure 1**). The extensive networking between these proteins indicates their involvement in similar pathways or processes.

A total of 668 neighbor proteins were retrieved for query proteins. They were colored in white and gray balls. White balls represent the proteins following the similar pathways as do the query proteins, whereas the gray balls represent the proteins following dissimilar different pathways. The query and neighbor proteins were observed to be following NGF, NT, adhesion, MAPK, and few more pathways.

We have hypothesized a network of proteins which is suggested to be the mechanism behind seizures (**Figure 2**). The interactions were later segregated into smaller routes to understand the flow of information inside the brain during a seizure (**Figure 3(a)–(d)**). The starting point for the current study of routes was finalized to be adenosine A1 receptor (ADORA1), while the last (effector molecule) was chosen to be gamma-aminobutyric acid B receptor 1 (GABBR1), we have limited our hypothesis between these two proteins.

4.1 Route 1

Adenosine is observed to be upregulated in TLE. Later it binds with ADORA1 and inhibits the signal transducer activity of GNAS (guanine nucleotide-binding protein G(s) subunit alpha isoforms XLas) protein [12]. Further, GNAS which stimulates the activity of adenylate cyclase type 5 (ADCY5) when inhibited leads to inhibition of ADCY5, and as a result there is no production of secondary messenger cAMP (**Figure 3(a)**) [13]. ADCY5 is also regulated by protein kinase C alpha type (PRKCA), and it inhibits its catalytic activity. YWHAZ then acts as an adaptor

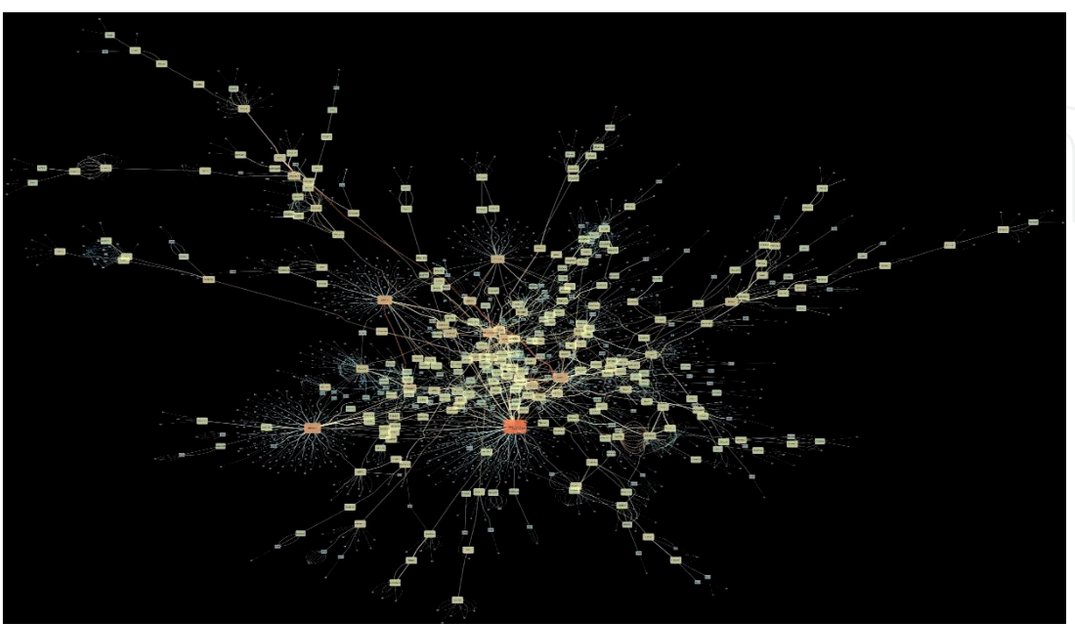

Figure 1.
The network generated by Cytoscapev2.7.2. The figure represents the master network, i.e., clusters of proteins communicating with each other during TLE conditions. It also reports the pathways mostly followed by the proteins involved in this network.

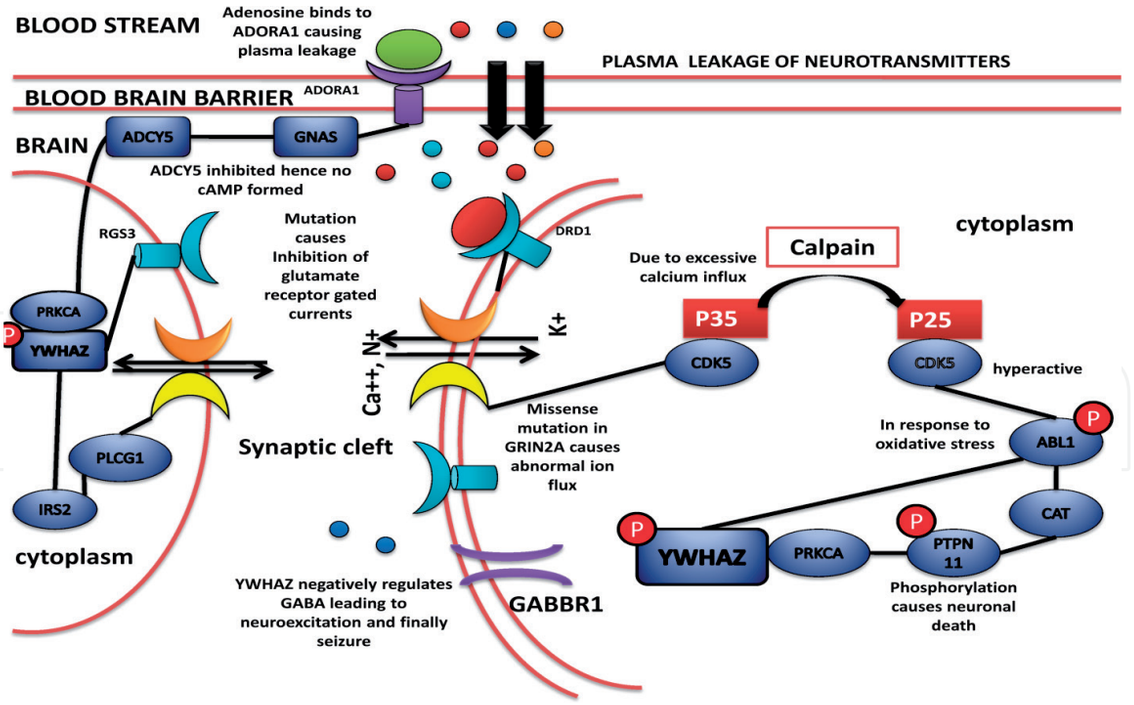

Figure 2.
Pictorial representation of seizure events in the brain during generalized seizures. The figure shows the detailed graphic representation of events occurring in the brain during focal seizure. Proteins involved in the hypothesis are shown in blue color. The process is redrawn as a simplified diagram for easy understanding.

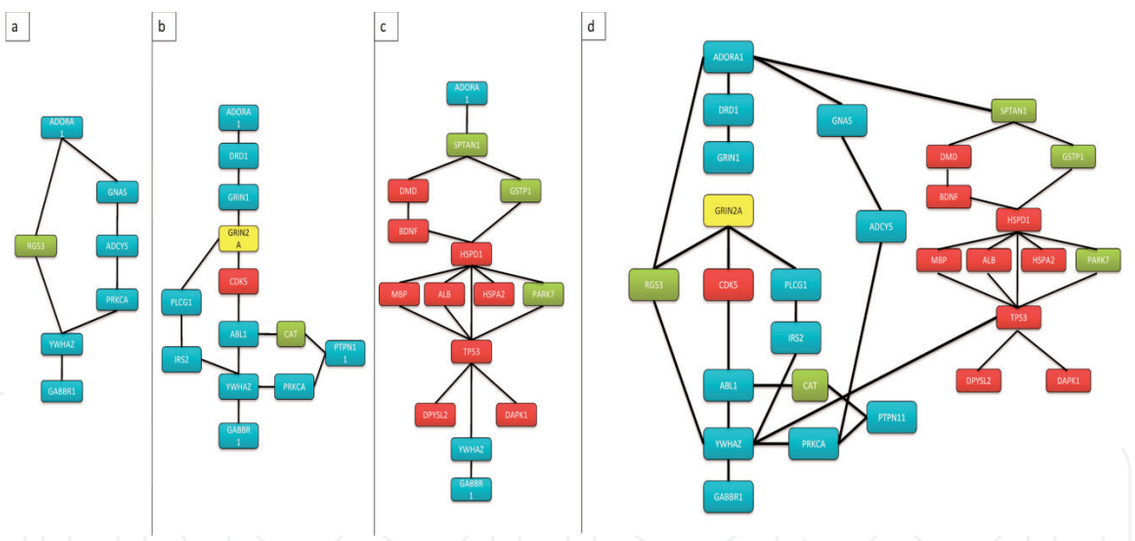

Figure 3.
The possible routes involved in a seizure: (a) route 1, (b) route 2, (c) route 3, and (d) simplified diagram of cumulative routes. The figure represents the different hypotheses of focal seizure generation via three different routes.

protein which interacts with PRKCA signaling molecule [14]. Ultimately YWHAZ negatively regulates GABA causing neuroexcitation, and hence seizure occurs [15].

Alternatively, ADORA1 is also inversely proportional to regulators of G protein signaling 3 (RGS3) which are G protein signaling molecules. RGS3 is downregulated by YWHAZ, causing no activation of MAP kinases which leads to altered cell growth and proliferation [16].

4.2 Route 2

Upregulation of adenosine in the body results in increased permeability of the BBB which leads to excessive influx of neurotransmitters [7]. Due to lesser reuptake

of these neurotransmitters by DRD1(dopamine receptor D1), it inhibits the activity of ADCY5 leading to decreased production of cAMP(secondary messenger) [17]. Coupling of DRD1 and glutamate receptor, ionotropic, N-methyl, D-aspartate 1 (GRIN1) causes inhibition of NMDA glutamate receptor-gated currents. GRIN 1 and glutamate receptor, ionotropic, N-methyl, D-aspartate 2A (GRIN 2A) are subunits of NMDA receptor [18]. Missense mutation in GRIN2A initiates phospholipase c, gamma 1 (PLCG1) [19]. Initiation of PLCG1 separates phosphatidylinositol 4, 5-bisphosphate (PIP2) into the second messengers, diacylglycerol (DAG) and inositol 1, 4, 5-trisphosphate (IP3), which play a vital role in intracellular signal transduction. An interruption in this signaling pathway causes altered synaptic transmission (**Figure 3(b)**) [19]. When mutated GRIN2A interacts with PLCG1, it causes hindrance in the production of secondary messengers and ultimately alters the intracellular signal transduction. Thereafter, PLCG1 does not activate, and no phosphorylation of tyrosine kinases takes place. It further causes no stimulation of insulin receptor substrate 2 (IRS2) [20]. This results in negative regulation of insulin which is facilitated by association of IRS2 and YWHAZ. Then, YWHAZ ultimately downregulates GABA, causing a seizure.

Alternatively, it is known that GRIN1 is also responsible for verbal memory and its inhibition causes speech impairment during a seizure (**Figure 3(b)**). GRIN2A is significant for neuronal activity and development. But a missense mutation of GRIN2A leads to abnormal neuronal ion flux and electrical transmission leading to developmental abnormalities [21]. GRIN1 and GRIN2A are the two subunits of NMDA glutamate receptor. Mutated GRIN2A causes excessive Ca^{2+} influx which causes conversion of P35 to P25 by calcium-dependent protease, calpain, during neurotoxicity and contributes to the pathological state. Elevation in levels of P25 leads to hyperactivation of cyclin-dependent kinase 5 (CDK5) which causes impairment in behavior, cognition, and synaptic plasticity [22]. Further elevated levels of CDK5 and oxidative stress phosphorylates ABL proto-oncogene 1, non-receptor tyrosine kinase (ABL-1) which is a ubiquitous non-receptor tyrosine kinase which promotes apoptosis and cytoskeleton remodeling [23]. ABL-1 in response to oxidative stress downregulates catalase (CAT) activity [24]. CAT binds actively to tyrosine-protein phosphatase non-receptor 11 (PTPN11) which is responsible for RAS/MAPK signaling which further leads to apoptosis. Low levels of CAT cause decreased resistance of PTPN11 toward hydrogen peroxide [25]. PTPN11 is phosphorylated by PRKCA, triggering neuronal death. YWHAZ acts as an adaptor protein which interacts with PRKCA signaling molecule. In response to DNA damage, ABL-1 is targeted into the nucleus, and it phosphorylates YWHAZ causing the activation of JNK signaling which is responsible for apoptosis [26].

4.3 Route 3

ADORA1 is activated by the upregulation of adenosine due to any external stimulus [27]. As a result, the permeability of BBB increases causing influx of calcium ions. Further, activation of ADORA1 causes downregulation of spectrin alpha chain, non-erythrocytic 1 (SPTAN1) which regulates the entry of calcium ions in neurons [27]. Due to decreased availability of calcium ions, DMD (dystrophin) is upregulated, and no proper synaptic transmission takes place [28]. Simultaneously, SPTAN1 interacts with glutathione S-transferase P (GSTP1) which is an antioxidant enzyme, regulating CDK5; downregulation of GSTP1 causes downregulation of CDK5; hence neurodegeneration initiates. On the other hand, DMD interacts with brain-derived neurotrophic factor (BDNF) which controls the neuronal excitability, is upregulated, and causes hyperexcitability [29]. BDNF and GSTP1 together interact with HSPD1 (60 kDa heat shock protein, mitochondrial),

which prevents misfolding of proteins, but it gets upregulated [30]. HSPD1 binds to five more proteins, namely, myelin basic protein (MBP), serum albumin (ALB), heat shock-related 70 kDa protein 2 (HSPA2), protein deglycase DJ-1 (PARK7), and cellular tumor antigen p53 (TP53). MBP forms the myelin sheath of a neuron, and its upregulation causes dysfunctioning of protein. The upregulation of ALB hinders its binding to the calcium ions which are leaked from the BBB and alter the osmotic pressure of neuronal cells [31]. Further HSPA2, which prevents aggregation of proteins, gets upregulated, it does not perform its function properly. PARK7 in response to increased oxidative stress does not perform its function of reducing the number of free radicals. The upregulation of TP53, a tumor suppressor protein, results in downregulation of apoptosis regulator Bcl-2 (BCL-2). BCL-2 induces apoptosis and causes neurodegeneration [32]. Further, TP53 interacts with YWHAZ which in response to oxidative stress arrests the cell cycle [33]. TP53 also interacts with dihydropyrimidinase-related protein 2 (DPYSL2) and death-associated protein kinase 1 (DAPK1). DPYSL2 modulates the growth of neurons which is upregulated and causes hindrance in transmission of electric signals, causing focal seizures. Normally, DAPK1 prevents autophagy in normal conditions, when the cell is under stress, it is elevated and causes destruction of neuronal cells [34].

From the abovementioned theories, the nearest and most realistic route for causing seizure generation is proposed to be route 3. This pathway was picked because it has those proteins which are differentially regulated and cause electrical irregularity in the brain, bringing on a focal seizure. Repetitive seizures cause weaknesses in cognizance, learning, and memory. Also, it has been seen the GABA levels were found to be low in brain tissue of epileptic patients. Real occasions during focal seizure are transformation of GRIN2A which brings about hindrance in neuronal particle flux. Our study concludes that the extended protein signal finally reaches to YWHAZ which causes hyperexcitation. This was due to inefficient binding of GABA to GABBR1 receptor (**Figure 2**).

5. Conclusion

The study hypothesized the series of protein-protein interactions which may potentially occur during a focal seizure. Out of the three plausible paths for the initiation of focal seizure, the most appropriate is route 3 which starts with upregulation of adenosine. Route 3 causes cell cycle arrest and starts the apoptosis of neuronal cells along with the hindrance in synaptic transmission [35].

Although AEDs have been formulated for numerous receptors, its generation is still an unresolved question. A study by Steinlein et al. showed that mutations in genes encoding for voltage-gated and ligand-gated ion channels like GABBR1 receptors and nicotinic acetylcholine receptors are mutated. These mutated receptors cause electrical imbalance in the brain [36].

Similarly, a study by Suls et al. suggested that some proteins undergo genetic mutations in TLE [37]. In the past research, it has been seen that mutations in GLUT1 causes epilepsy. GLUT1 is the major receptor for gluten molecule. GLUT1 mutation imbalances the concentration of gluten, K^+, and albumin across the BBB and impairs the K^+ buffering in the brain. Gluten metabolism and transport is also disturbed which becomes the major reason for neuronal excitability [38]. Some studies also suggested that seizure is caused due to mutation in CDK5 [39]. The modulation of GABAergic inhibition by NMDA receptors may cause the synaptic plasticity [40].

On the other hand, excessive influx of calcium ions leads to hindered synaptic transmission [35], due to which heat shock proteins are unable to perform their

normal function. In response to oxidative stress proteins like BDNF, MBP, ALB, and TP53, DPYSL2 gets upregulated and induces apoptosis, hence causing neurodegeneration [22]. Then, YWHAZ finally binds to GABBR1 (helps in preventing ion leakage on the membrane and neuronal hyperactivity) and makes it unable to bind to GABA, which is an inhibitory neurotransmitter. Therefore, YWHAZ can be studied in future as a drug target for epileptic focal seizures.

To date, there were just a couple of protein receptors targeted for antiepileptic drugs which manage the pathology. Also, there are no medications to prevent a seizure, and no such receptor has been predicted for the same. Therefore, the present study proposes that the proteins involved in route 3 may give some understanding into the fundamental mechanism behind seizures. Protein that can be focused for designing AEDs can be YWHAZ.

Acknowledgements

We would like to acknowledge Dr. Ashok K. Chauhan, Founder President, Amity University Uttar Pradesh, Noida, for providing the infrastructure and support.

Author details

Ruchi Jakhmola Mani and Deepshikha Pande Katare*
Proteomics and Translational Research Lab, Centre for Medical Biotechnology, Amity Institute of Biotechnology, Amity University, Noida, India

*Address all correspondence to: dpkatare@amity.edu

References

[1] Banerjee PN, Filippi D, Hauser WA. The descriptive epidemiology of epilepsy—A review. Epilepsy Research. 2009;**85**:31-45

[2] Schiffer RB, Babigian HM. Behavioral disorders in multiple sclerosis, temporal lobe epilepsy, and amyotrophic lateral sclerosis: An epidemiologic study. Archives of Neurology. 1984;**41**:1067-1069

[3] Haneef Z, Lenartowicz A, Yeh HJ, et al. Functional connectivity of hippocampal networks in temporal lobe epilepsy. Epilepsia. 2014;**55**:137-145

[4] Clancy MJ, Clarke MC, Connor DJ, et al. The prevalence of psychosis in epilepsy; a systematic review and meta-analysis. BMC Psychiatry. 2014;**14**:1

[5] Rigau V, Morin M, Rousset MC, et al. Angiogenesis is associated with blood–brain barrier permeability in temporal lobe epilepsy. Brain. 2007;**130**:1942-1956

[6] Bedner P et al. Astrocyte uncoupling as a cause of human temporal lobe epilepsy. Brain. 2015;**138**(5):1208-1222

[7] Lapointe E, Deacon C, Bocti C, et al. Temporal lobe atrophy: Frequent in elderly with epilepsy of unknown etiology. Clinical Neurophysiology. 2016;**127**:e161

[8] Ben-Ari Y, Khalilov I, Kahle KT, et al. The GABA excitatory/inhibitory shift in brain maturation and neurological disorders. The Neuroscientist. 2012;**18**:467-486

[9] Briggs SW, Galanopoulou AS. Altered GABA signaling in early life epilepsies. Neural Plasticity. 2011;**2011**. Article ID 527605

[10] Jacob TC, Moss SJ, Jurd R. GABAA receptor trafficking and its role in the dynamic modulation of neuronal inhibition. Nature Reviews. Neuroscience. 2008;**9**:331-343

[11] Sebba J, Hogg LLJ. Profound Retardation and Multiple Impairment: Volume 3: Medical and Physical Care and Management. Springer; 1 Dec 2013

[12] Baraldi PG, Tabrizi MA, Fruttarolo F, et al. Recent improvements in the development of A2B adenosine receptor agonists. Purinergic Signal. 2008;**4**:287-303

[13] Hampton M, Melvin RG, Andrews MT. Transcriptomic analysis of brown adipose tissue across the physiological extremes of natural hibernation. PLOS One. 2013;**8**:e85157

[14] Gannon-Murakami L, Murakami K. Selective association of protein kinase C with 14-3-3 ζ in neuronally differentiated PC12 cells stimulatory and inhibitory effect of 14-3-3 ζ in vivo. The Journal of Biological Chemistry. 2002;**277**:23116-23122

[15] Couve A, Kittler JT, Uren JM, et al. Association of GABA B receptors and members of the 14-3-3 family of signaling proteins. Molecular and Cellular Neurosciences. 2001;**17**:317-328

[16] Kleppe R, Rosati S, Jorge-Finnigan A, et al. Phosphorylation dependence and stoichiometry of the complex formed by tyrosine hydroxylase and 14-3-3γ. Molecular & Cellular Proteomics. 2014;**13**:2017-2030

[17] Levy F, de Leon J. Dopamine ADHD/OCD theories: Is glutamine part of the story? Neurotransmitter. 2015;**2**

[18] Petralia RS, Al-Hallaq RA, Wenthold RJ. Trafficking and targeting

of NMDA receptors. Biology of the NMDA Receptor. 2009;**1**:149-200

[19] Jang HJ, Yang YR, Kim JK, et al. Phospholipase C-γ1 involved in brain disorders. Advances in Biological Regulation. 2013;**53**:51-62

[20] Ghorbani S, Fossbakk A, Jorge-Finnigan A, et al. Regulation of tyrosine hydroxylase is preserved across different homo-and heterodimeric 14-3-3 proteins. Amino Acids. 2016;**48**:1221-1229

[21] Clasadonte J, Dong J, Hines DJ, et al. Astrocyte control of synaptic NMDA receptors contributes to the progressive development of temporal lobe epilepsy. Proceedings of the National Academy of Sciences. 2013;**110**:17540-17545

[22] Hawasli AH, Bibb JA. Alternative roles for Cdk5 in learning and synaptic plasticity. Biotechnology Journal. 2007;**2**:941-948

[23] Chen L, Wang Z, Tang B, et al. Altered expression of c-Abl in patients with epilepsy and in a rat model. Synapse. 2014;**68**:306-316

[24] Cao C, Leng Y, Kufe D. Catalase activity is regulated by c-Abl and Arg in the oxidative stress response. The Journal of Biological Chemistry. 2003;**278**:29667-29675

[25] Yano S, Arroyo N, Yano N. SHP2 binds catalase and acquires a hydrogen peroxide-resistant phosphatase activity via integrin-signaling. FEBS Letters. 2004;**577**:327-332

[26] Yoshida K, Yamaguchi T, Natsume T, et al. JNK phosphorylation of 14-3-3 proteins regulates nuclear targeting of c-Abl in the apoptotic response to DNA damage. Nature Cell Biology. 2005;**7**:278-285

[27] da Silva Fernandes MJ, Amorim RP, Carneiro JE, et al. Proteomics-based strategy to identify biomarkers and pharmacological targets in temporal lobe epilepsy. In: Pharmacoresistance in Epilepsy. New York: Springer; 2013. pp. 115-126

[28] Lee TS, Eid T, Mane S, et al. Aquaporin-4 is increased in the sclerotic hippocampus in human temporal lobe epilepsy. Acta Neuropathologica. 2004;**108**:493-502

[29] Sadakata T, Shinoda Y, Oka M, et al. Reduced axonal localization of a Caps2 splice variant impairs axonal release of BDNF and causes autistic-like behavior in mice. Proceedings of the National Academy of Sciences. 2012;**109**:21104-21109

[30] Gammazza AM, Colangeli R, Orban G, et al. Hsp60 response in experimental and human temporal lobe epilepsy. Scientific Reports. 2015;**5**

[31] Van Vliet EA, da Costa AS, Redeker S, et al. Blood–brain barrier leakage may lead to progression of temporal lobe epilepsy. Brain. 2007;**130**:521-534

[32] Kilany A, Raouf ER, Gaber AA, et al. Elevated serum Bcl-2 in children with temporal lobe epilepsy. Seizure. 2012;**21**:250-253

[33] Waterman MJ, Stavridi ES, Waterman JL, et al. ATM-dependent activation of p53 involves dephosphorylation and association with 14-3-3 proteins. Nature Genetics. 1998;**19**:175-178

[34] Hainsworth AH, Allsopp RC, Jim A, et al. Death-associated protein kinase (DAPK1) in cerebral cortex of late-onset Alzheimer's disease patients and aged controls. Neuropathology and Applied Neurobiology. 2010;**36**:17-24

[35] Voss LJ, Jacobson G, Sleigh JW. Bridging the Gap—Understanding the Role of Gap Junctions in Seizures. IntechOpen Access Publisher; 2011

[36] White HS, Smith MD, Wilcox KS. Mechanisms of action of antiepileptic drugs. International Review of Neurobiology. 2007;**81**:85-110

[37] Scheffer IE, Berkovic SF. The genetics of human epilepsy. Trends in Pharmacological Sciences. 2003;**24**:428-433

[38] Suls A, Mullen SA, Weber YG, et al. Early-onset absence epilepsy caused by mutations in the glucose transporter GLUT1. Annals of Neurology. 2009;**66**:415-419

[39] Castrén M, Gaily E, Tengström C, et al. Epilepsy caused by CDKL5 mutations. European Journal of Paediatric Neurology. 2011;**15**(1):65-69

[40] Stelzer A, Slater NT, Bruggencate GT. Activation of NMDA receptors blocks GABAergic inhibition in an in vitro model of epilepsy. Nature. 1987;**326**(6114):698-701