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REVIEW ARTICLE

The Anticonvulsant Effects of Ketogenic Diet on Epileptic Seizures and Potential Mechanisms

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Abstract: ***Background*:** Epilepsy is a syndrome of brain dysfunction induced by the aberrant excitability of certain neurons. Despite advances in surgical technique and anti-epileptic drug in recent years, recurrent epileptic seizures remain intractable and lead to a serious morbidity in the world. The ketogenic diet refers to a high-fat, low-carbohydrate and adequate-protein diet. Currently, its beneficial effects on epileptic seizure reduction have been well established. However, the detailed mechanisms underlying the anti-epileptic effects of ketogenic diet are still poorly understood. In this article, the possible roles of ketogenic diet on epilepsy were discussed.

***Methods*:** Data was obtained from the websites including Web of Science, Medline, Pubmed, Scopus, based on these keywords: "Ketogenic diet" and "epilepsy".

***Results*:** As shown in both clinical and basic studies, the therapeutic effects of ketogenic diet might involve neuronal metabolism, neurotransmitter function, neuronal membrane potential and neuron protection against ROS.

***Conclusion*:** In this review, we systematically reviewed the effects and possible mechanisms of ketogenic diet on epilepsy, which may optimize the therapeutic strategies against epilepsy.

Keywords: Epilepsy, seizure, ketogenic diet, GABA, neuron, animal.

1. INTRODUCTION

Epilepsy is a group of neurological disorders characterized by a long term risk of recurrent seizures [1], which vary from brief and nearly undetectable to long periods of vigorous shaking, causing physical injuries including bone fractures and bite to tongue tips occasionally. These epileptic seizures may present in several ways depending on the area of the brain involved [2]. According to the International League Against Epilepsy (ILAE) statistics, as of 2013, epilepsy jeopardized approximately 22 million people, accounting for about 116,000 deaths [3, 4]. Furthermore, epilepsy became more prevalent as people age, causing a great burden to governments worldwide [5]. Although in certain cases, epilepsy results from birth defect, cerebral inflammation or tumor, the cause of most cases remains unknown. Genetic mutations and homozygous variants were only identified in a small proportion of epilepsy cases [6]. Some of the patients might be candidates for surgical resection of abnormal tissue, but this option is simply available for those with such lesions. Today, the mainstay treatment of epilepsy is anticonvulsant medications [7]. About 30% of people, however, continue to have seizures despite anticonvulsant drug treatment [8].

The ketogenic diet (KD) refers to a high-fat, low-carbohydrate and adequate-protein diet, including various recipes. The classical KD consists of long-chain triglycerides (LCTs), usually applied in KD ratio of 4:1 or 3:1 for fat: nonfat (protein and carbohydrates). Ketogenic diet therapies for epilepsy were initially introduced in scientific literature since the early 1900s. In spite of the development of newer drugs and other treatment, ketogenic dietary therapy for epilepsy is recently emerging as a non-pharmacological alternative treatment option for physicians and patients, especially in whom resection surgery is not feasible [9]. A recent Cochrane review has shown that at least 38% of the patients on the KD obtain a 50% reduction in seizures as compared to controls at 3 months [10]. Multiple research groups have reported that KD is a safe and effective treatment for childhood epilepsies, especially for infantile epilepsies. As described in a randomized controlled trial of KD, the classic KD therapy appears to decrease the number of seizures by half in about 30–40% of children [9]. Therefore, KD serves as an established treatment for drug-resistant childhood epilepsy and a reasonable option in those who have epilepsy that is not improved with medications or for those where

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surgery is not available. DM and colleagues performed 4 months of a randomized controlled study in children and adolescents by Follow-up assessments to investigate its effects of cognition and aspects of behavior. Their study showed a positive impact of the KD on behavioral and cognitive functioning in children and adolescents with refractory epilepsy. More specifically, an activated mood and cognitive activation were observed in patients treated with the KD [11].

Side effects of KD include stomach and intestinal problems in 30%, and there are long term concerns about cardiac diseases [9]. In clinical practice, carrying on the classic KD diet in pediatric patients is not always easy, due to its relatively strict dietary regimen which might lead to severe side effects [12]. Although the extent of ketosis is not always proportionately correlated with better epileptic seizure treatment during KD therapy, a consistent and high level of serum ketosis is critical to control epileptic seizure. To overcome such disadvantages, a more balanced and easily accepted alternative dietary therapy in the form of modified Atkins diet (MAD) has been widely introduced in the past decade [13]. According to a number of pilot studies, the MAD is as effective as the classic KD for recurrent epileptic seizures [14, 15]. Kim and her colleagues have found that the MAD dietary regimens showed great advantages with respect to better tolerability and fewer side effects, thus it can be considered as the primary option for the treatment of intractable epilepsy in children [16]. In addition, the MAD protocol combined with calorie restriction might improve seizure outcomes in epilepsy patients [17]. However, as the mechanisms through which KD confers to its anti-epileptic effects remain unknown, the clinical guidance of KD treatment remains controversial.

In this article, we will discuss the potential mechanisms underlying the therapeutic effects of KD on recurrent epilepsy by presenting important experimental evidences and hypothesis, aiming to explore possible biological basis behind such efficacies and provide new insight to the KD treatment in recurrent epilepsy.

2. ANTICONVULSANT EFFECTS OF KETONE BODIES ON EPILEPSY

To better understand the therapeutic effects of KD on epilepsy treatment and its molecular basis, many laboratories have focused on the anticonvulsant effects of different ketone bodies within different pharmacological doses. Ketone bodies, consisting of acetoacetate and β-hydroxybutyrate are derived from fat catabolism in liver mitochondria and their concentration in blood is inversely related to that of glucose. β-hydroxybutyrate is the predominant blood ketone body and is oxidized to acetoacetate in the mitochondria before entering the TCA cycle. Rho and colleagues found that it was acetoacetate and acetonerather than β-hydroxybutyrate that can significantly decreased the epileptic seizures in the seizure-susceptible juvenile mouse model, indicating anticonvulsant effects of these ketone bodies [18]. In addition, rapid administration of acetone through intraperitoneal administration reduced seizure susceptibility in a series of seizure tests, such as the maximal electroshock test and the amygdale kindling test [19]. Gaisor and colleagues demonstrated that acetone, *per se*, protected the pentylenetetrazol and 4- aminopyridine induced epileptic seizures in juvenile mouse model, which further supported its anticonvulsant effects [20, 21]. In epilepsy patients, however, the serum levels of acetone during KD were extremely lower than the levels expected to be therapeutic [22], suggesting that acetone alone might be insufficient to account for the therapeutic roles of KD in epilepsy.

To investigate the effects of KD and anticonvulsant drug combination, Szot and his colleagues performed co-administration of valproate and ketogenic diet and examined its effect. In majority of the cases, it seemed safe to co-administrate valproate and ketogenic diet. Only in two cases, valproate made a negative influence on ketosis. Their research sheds new light on the combination strategy of KD and anticonvulsant drugs, which is a worthy future study [23].

3. POSSIBLE MECHANISMS UNDERLYING KD TREATMENT IN EPILEPSY

3.1. The Effects of KD on Neuron Metabolism

In normal condition, brain electrical activity maintains non-synchronous. Its activity is modulated by certain factors both within the neuron and the cellular environment. Although the anticonvulsant mechanisms of the KD remain unclarified, many research efforts have proposed several important theories and hypothesis to explain the decreased seizure threshold during KD treatment. One suggested mechanism is the effect of KD on energy metabolism to reduce epileptic seizures. Despite that glucose is the preferred energy substrate. The brain metabolizes ketone bodies for energy when glucose levels are rapidly decreased during KD. It has been established that the efficacy of the KD for managing epilepsy is best when the diet is administered following a fast or when total calories are restricted. Furthermore, the seizure protective effects of the KD are found to rely on the maintenance of low blood glucose levels [24]. Reduced blood glucose levels force the brain to burn ketones for energy. Ketone metabolism gradually reduces neuronal excitability, thereby producing effects on neurotransmitter levels and neuron membrane potential. Thus, a restriction of caloric energy intake should enhance the antiepileptic effects of the KD.

Under normal physiological conditions, the brain derives almost all of its energy from the aerobic oxidation of glucose. Glucose transporters (GLUT-1) are abundant in the brain capillary endothelial cells and facilitate the diffusion of glucose through the blood-brain barrier. It has been established that epilepsy is caused by excessive and abnormal nerve cell activity in the cortex of the brain [25]. Thus, epileptic seizures are the result of excessive and abnormal nerve cell activity in the cortex of the brain [4]. Brain glycolysis can be activated by neuronal activity and blocked by citrate and ATP synthesized through the TCA cycle and oxidative phosphorylation. Glucose uptake and metabolism were found to be highly accelerated during epileptic seizures than during normal condition and neuronal excitability, and epileptic seizures are directly related to rapid glucose utilization and glycolysis. Cheng and colleagues found that IGF1R and

GLUT expressions were significantly upregulated during KD, suggesting that IGF system and glucose transporters might participate in ketogenic diet-induced seizure suppression [26]. Glut1 is the most relevant glucose transporter in the brain. Notably, all Glut1 syndromes respond well to a ketogenic diet (KD) and most of the patients show a rapid seizure control. To further investigate its role in KD treatment, Becker's team sequenced SLC2A1 that encoding Glut1, in a cohort of 28 patients with different forms of pharmaco-resistant epilepsies responding well to KD. However, no mutations in SLC2A1 were determined [27]. The association between therapeutic effects of KD and Glut1 induced glucose metabolism is still needed to investigate. Qian's research team demonstrated that the status of metabotropic glutamate receptors (mGluRs) and their downstream signal pathways were correlated with epileptogenesis, suggesting that glutamate metabolism might also regulate epileptic seizures [28]. However, there is no direct evidence indicating the correlation between glutamate metabolism and the therapeutic effects of KD.

In liver and brain, mitochondrial 3-hydroxy-3-methylglutaryl-CoA synthase (mHS) is the critical enzyme of ketogenesis. Cullingford and colleagues detected the gene expression profile among the rats with different diet recipes including a calorie-restricted ketogenic diet (KCR) and a normal diet ad libitum (NAL). As a result, they found that KCR-fed rats exhibited a significant 2-fold increase in both liver and brain mHS mRNA as compared to NAL-fed rats [29]. Their data indicated that mHS expression might play a role in KD treatment. Its downstream molecular basis is required to be elucidated in the future studies. Fraser and colleagues found that most polyunsaturated fatty acids (PUFA) were greatly elevated in children within KD and the increased arachidonate was significantly correlated with improved seizure control, suggesting PUFA may represent a critical anticonvulsant mechanism of the KD [30]. Pyruvate dehydrogenase complex (PDC) is essential for the neuron development and its deficiency can block acetyl-CoA and ATP synthesis, leading to lactic acidosis, neuroanatomic defects, developmental delay, and early death. In an earlier study, Wexler and colleagues found that Pyruvate dehydrogenase complex (PDC) deficiency was improved by the KD with varying degrees of carbohydrate restriction [31]. In addition, a homozygous pathogenic variant of MED23 was recently identified in an epilepsy patient, who showed a complete response to the KD treatment. As MED23 has been established as a regulator of energy homeostasis and glucose production, it was proposed that the reduced glucose level induced by MED23 dysfunction can be treated by energy substitutes of ketone boides [32]. Altogether, these studies indicated that the therapeutic effects of KD in epilepsy might result from increased ketone bodies as energy source and bypassing glucose metabolism.

3.2. The Effects of KD on Neurotransmitter Function

GABA signaling is the most well studied target of investigation since mouse models of epilepsy induced by GABA antagonists exhibited a remarkable response to KD treatment [33]. Moreover, in rat synaptosomes GABA synthesis was greatly increased and maintained at a high level by ketone bodies, which may contribute to the beneficial effect of a ketogenic diet in the treatment of epilepsy [34]. Many clinical studies showed increased GABA levels in the cerebrospinal fluid of patients on a ketogenic diet [35, 36], further supporting that GABA might be regulated by ketone bodies.

Aspartate is an inhibitor of glutamate decarboxylase, which catalyzes α-ketoglutarate to GABA. Consequently, a decrease in aspartate may promote the synthesis of GABA. Yudkoff's group proposed a promising hypothesis that during KD, decreased aspartate levels induced by ketone bodies can facilitate the conversion of excitatory neurotransmitter glutamate to glutamine in astrocytes. Meanwhile, the inhibitory neurotransmitter glutamine is then taken up into neurons and eventually converted to GABA, making an inhibitory influence on neuronal activities [37]. However, some other studies found that there is no change in GABA levels in KD fed rodents [38], suggesting GABA levels in different brain area as well as other factors of GABA function are also important for KD treatment.

Except for the GABA levels, Szot and colleagues reported that functional noradrenergic nervous system is also required for the KD to exert an anticonvulsant effect [39]. Consistent with their results, Masino's group found that KD can reduce seizures in mice by increasing activation of adenosine A1 receptors (A1Rs). The KD reduced adenosine kinase, the major adenosine-metabolizing enzyme. Hippocampal tissue resected from patients with medically intractable epilepsy demonstrated increased adenosine kinase, suggesting that adenosine deficiency may be relevant to human epilepsy and that KD can reduce seizures by increasing A1R-mediated inhibition [40]. However, such results are needed to be further confirmed through clinical investigation.

3.3. The Effects of KD on Neuronal Membrane

Another potential mechanism underlying the effects of ketone bodies on epileptic seizures is their effects on neuronal membrane transporters. Ketone bodies may alter the behavior of vesicular glutamate transporters (VGLUTs) that are responsible for filling pre-synaptic vesicles with glutamate in a Cl^-dependent manner. Juge and colleagues demonstrated that Cl^- is an allosteric activator of VGLUTs that is competitively inhibited by ketone bodies (acetoacetate) more strongly than β-hydroxybutyrate [41]. K_{ATP} channels play a key role in CA3 hippocampal neuron hyperpolarization when the neurons were cultured in low glucose media [42]. In cultured mouse hippocampal dentate gyrus neurons, β-hydroxybutyrate increased K_{ATP} channel opening, which could partially explain the effects of KD treatment [43]. However, the exact relevance between the pharmacological doses of ketone bodies and the activities of VGLUTS and K_{ATP} channels remain unknown.

Abnormal gap junctional communication is an underlying mechanism involved in the generation and maintenance of seizures. Consequently, the effects of gap junction blockers were determined in seizure models. As a result, these gap junction blockers can reduce both amplitude and frequency of the epileptiform activity, and modify the behavioral parameters related to seizures *in vivo* assay [44]. However, no

clinical evidence supported the correlation between KD treatment and the gap junctional communication.

3.4. The Effects of KD on Neuron Protection Against ROS

According to the Maalouf's study, reactive oxygen species production resulting from glutamate exposure can be reversed by ketone bodies in primary cultures of rat neocortical neurons through NADH oxidation [45]. The KD also promotes the biosynthesis of glutathione *via* the transcription factorNrf2, regulating the neuronal ROS levels [46]. Ketone bodies also protect against cell death through reducing mitochondrial ROS production after neocortical slices are exposed to hydrogen peroxide [47]. These experimental data suggest that ketone bodies may be neuroprotective against reactive oxygen species produced in neuropathological states. However, the effects of ROS alteration induced by KD on epileptic seizures need both of experimental and clinical data.

CONCLUSION

The antiepileptic efficacy of KD in epilepsy has been found to increase the number of experimental evidences and raise a promising therapeutic strategy against epilepsy, due to which molecular mechanism remains poorly studied. Intense research efforts in neurology over the past decade have provided significant insights into the molecular basis of KD treatment in epilepsy. To date, the exact mechanism how the brain shifts into the activity of a seizure with its excessive synchronization remains poorly understood. Supported by certain clinical and preclinical studies, as aforementioned, the therapeutic effects of KD might be based on the molecular basis that involves neuronal metabolism, neurotransmitter function, neuronal membrane potential and neuron protection against ROS. However, more detailed mechanisms are still to be elucidated in future studies not only to optimize the clinical application of KD treatment in epilepsy, but also to develop novel drug targets and therapeutic strategies.

CONSENT FOR PUBLICATION

Not applicable.

CONFLICT OF INTEREST

The authors declare no conflict of interest, financial or otherwise.

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REFERENCES

[1] Liu, W.; Ge, T.; Pan, Z.; Leng, Y.; Lv, J.; Li, B. The effects of herbal medicine on epilepsy. *Oncotarget*, **2017**, *8*(29), 48385-48397.

[2] Nazıroğlu, M. TRPV1 Channel: A potential drug target for treating epilepsy. *Curr. Neuropharmacol.,* **2015**, *13*(2), 239-247. [http://dx.doi.org/10.2174/1570159X13666150216222543] [PMID: 26411767]

[3] Celli, R.; Santolini, I.; Guiducci, M.; van Luijtelaar, G.; Parisi, P.; Striano, P.; Gradini, R.; Battaglia, G.; Ngomba, R.T.; Nicoletti, F. The α2δ subunit and absence epilepsy: Beyond calcium channels? *Curr. Neuropharmacol*., **2017**, *15*(6), 918-925.

[4] Mortality, G.B. Global, regional, and national age-sex specific all-cause and cause-specific mortality for 240 causes of death, 1990-2013: a systematic analysis for the Global Burden of Disease Study 2013. *Lancet,* **2015**, *385*(9963), 117-171. [http://dx.doi.org/10.1016/S0140-6736(14)61682-2] [PMID: 25530442]

[5] Brodie, M.J.; Elder, A.T.; Kwan, P. Epilepsy in later life. *Lancet Neurol.,* **2009**, *8*(11), 1019-1030. [http://dx.doi.org/10.1016/S1474-4422(09)70240-6] [PMID: 19800848]

[6] Pandolfo, M. Genetics of epilepsy. *Semin. Neurol.,* **2011**, *31*(5), 506-518. [http://dx.doi.org/10.1055/s-0031-1299789] [PMID: 22266888]

[7] Frey, H.H. Anticonvulsant drugs used in the treatment of epilepsy. *Probl. Vet. Med.,* **1989**, *1*(4), 558-577. [PMID: 2520134]

[8] Eadie, M.J. Shortcomings in the current treatment of epilepsy. *Expert Rev. Neurother.,* **2012**, *12*(12), 1419-1427. [http://dx.doi.org/10.1586/ern.12.129] [PMID: 23237349]

[9] Zhang, Y.; Liu, C.; Zhao, Y.; Zhang, X.; Li, B.; Cui, R.; Zhang, Y.; Liu, C.; Zhao, Y.; Zhang, X.; Li, B.; Cui, R. The effects of calorie restriction in depression and potential mechanisms. *Curr. Neuropharmacol*., **2015**, *13*(4), 536-42.

[10] Levy, R.G.; Cooper, P.N.; Giri, P. Ketogenic diet and other dietary treatments for epilepsy. *Cochrane Database Syst. Rev.,* **2012**, (3), CD001903. [PMID: 22419282]

[11] IJff, D.M.; Postulart, D.; Lambrechts, D.A.; Majoie, M.H.; de Kinderen, R.J.; Hendriksen, J.G.; Evers, S.M.; Aldenkamp, A.P. Cognitive and behavioral impact of the ketogenic diet in children and adolescents with refractory epilepsy: A randomized controlled trial. *Epilepsy Behav.,* **2016**, *60*, 153-157. [http://dx.doi.org/10.1016/j.yebeh.2016.04.033] [PMID: 27206235]

[12] Kang, H.C.; Chung, D.E.; Kim, D.W.; Kim, H.D. Early- and late-onset complications of the ketogenic diet for intractable epilepsy. *Epilepsia,* **2004**, *45*(9), 1116-1123. [http://dx.doi.org/10.1111/j.0013-9580.2004.10004.x] [PMID: 15329077]

[13] Sharma, S.; Sankhyan, N.; Gulati, S.; Agarwala, A. Use of the modified Atkins diet for treatment of refractory childhood epilepsy: a randomized controlled trial. *Epilepsia,* **2013**, *54*(3), 481-486. [http://dx.doi.org/10.1111/epi.12069] [PMID: 23294191]

[14] Kossoff, E.H.; Rowley, H.; Sinha, S.R.; Vining, E.P. A prospective study of the modified Atkins diet for intractable epilepsy in adults. *Epilepsia,* **2008**, *49*(2), 316-319. [http://dx.doi.org/10.1111/j.1528-1167.2007.01256.x] [PMID: 17919301]

[15] Miranda, M.J.; Mortensen, M.; Povlsen, J.H.; Nielsen, H.; Beniczky, S. Danish study of a modified Atkins diet for medically intractable epilepsy in children: can we achieve the same results as with the classical ketogenic diet? *Seizure,* **2011**, *20*(2), 151-155. [http://dx.doi.org/10.1016/j.seizure.2010.11.010] [PMID: 21126887]

[16] Kim, J.A.; Yoon, J.R.; Lee, E.J.; Lee, J.S.; Kim, J.T.; Kim, H.D.; Kang, H.C. Efficacy of the classic ketogenic and the modified Atkins diets in refractory childhood epilepsy. *Epilepsia,* **2016**, *57*(1), 51-58. [http://dx.doi.org/10.1111/epi.13256] [PMID: 26662710]

[17] Kim, Y.M.; Vaidya, V.V.; Khusainov, T.; Kim, H.D.; Kim, S.H.; Lee, E.J.; Lee, Y.M.; Lee, J.S.; Kang, H.C. Various indications for a modified Atkins diet in intractable childhood epilepsy. *Brain Dev.,* **2012**, *34*(7), 570-575. [http://dx.doi.org/10.1016/j.braindev.2011.09.013] [PMID: 22001499]

[18] Rho, J.M.; Anderson, G.D.; Donevan, S.D.; White, H.S. Acetoacetate, acetone, and dibenzylamine (a contaminant in l-(+)-beta-hydroxybutyrate) exhibit direct anticonvulsant actions *in vivo*. *Epilepsia,* **2002**, *43*(4), 358-361. [http://dx.doi.org/10.1046/j.1528-1157.2002.47901.x] [PMID: 11952765]

[19] Likhodii, S.S.; Serbanescu, I.; Cortez, M.A.; Murphy, P.; Snead, O.C., III; Burnham, W.M. Anticonvulsant properties of acetone, a brain ketone elevated by the ketogenic diet. *Ann. Neurol.,* **2003**, *54*(2), 219-226. [http://dx.doi.org/10.1002/ana.10634] [PMID: 12891674]

[20] Gasior, M.; Hartman, A.L.; Rogawski, M.A. The anticonvulsant activity of acetone does not depend upon its metabolites. *Epilepsia,* **2008**, *49*(5), 936-937. [http://dx.doi.org/10.1111/j.1528-1167.2007.01518_3.x] [PMID: 18454785]

[21] Gasior, M.; French, A.; Joy, M.T.; Tang, R.S.; Hartman, A.L.; Rogawski, M.A. The anticonvulsant activity of acetone, the major ketone body in the ketogenic diet, is not dependent on its metabolites acetol, 1,2-propanediol, methylglyoxal, or pyruvic acid. *Epi-*

lepsia, **2007**, *48*(4), 793-800. [http://dx.doi.org/10.1111/j.1528-1167.2007.01026.x] [PMID: 17386058]

[22] Seymour, K.J.; Bluml, S.; Sutherling, J.; Sutherling, W.; Ross, B.D. Identification of cerebral acetone by 1H-MRS in patients with epilepsy controlled by ketogenic diet. *MAGMA,* **1999**, *8*(1), 33-42. [PMID: 10383091]

[23] Spilioti, M.; Pavlou, E.; Gogou, M.; Katsanika, I.; Papadopoulou-Alataki, E.; Grafakou, O.; Gkampeta, A.; Dinopoulos, A.; Evangeliou, A. Valproate effect on ketosis in children under ketogenic diet. *Eur. J. Paediatr. Neurol.,* **2016**, *20*(4), 555-559. [http://dx.doi.org/10.1016/j.ejpn.2016.04.003] [PMID: 27117552]

[24] Greene, A.E.; Todorova, M.T.; Seyfried, T.N. Perspectives on the metabolic management of epilepsy through dietary reduction of glucose and elevation of ketone bodies. *J. Neurochem.,* **2003**, *86*(3), 529-537. [http://dx.doi.org/10.1046/j.1471-4159.2003.01862.x] [PMID: 12859666]

[25] Fisher, R.S.; van Emde, B.W.; Blume, W.; Elger, C.; Genton, P.; Lee, P.; Engel, J., Jr. Epileptic seizures and epilepsy: definitions proposed by the international league against epilepsy (ILAE) and the international bureau for epilepsy (IBE). *Epilepsia,* **2005**, *46*(4), 470-472. [http://dx.doi.org/10.1111/j.0013-9580.2005.66104.x] [PMID: 15816939]

[26] Cheng, C.M.; Kelley, B.; Wang, J.; Strauss, D.; Eagles, D.A.; Bondy, C.A. A ketogenic diet increases brain insulin-like growth factor receptor and glucose transporter gene expression. *Endocrinology,* **2003**, *144*(6), 2676-2682. [http://dx.doi.org/10.1210/en.2002-0057] [PMID: 12746332]

[27] Becker, F.; Schubert, J.; Weckhuysen, S.; Suls, A.; Grüninger, S.; Korn-Merker, E.; Hofmann-Peters, A.; Sperner, J.; Cross, H.; Hallmann, K.; Elger, C.E.; Kunz, W.S.; Madeleyen, R.; Lerche, H.; Weber, Y.G. Do Glut1 (glucose transporter type 1) defects exist in epilepsy patients responding to a ketogenic diet? *Epilepsy Res.,* **2015**, *114*, 47-51. [http://dx.doi.org/10.1016/j.eplepsyres.2015.04.012] [PMID: 26088884]

[28] Qian, F.; Tang, F.R. Metabotropic glutamate receptors and interacting proteins in epileptogenesis. *Curr. Neuropharmacol.,* **2016**, *14*(5), 551-562. [http://dx.doi.org/10.2174/1570159X14666160331142228] [PMID: 27030135]

[29] Cullingford, T.E.; Eagles, D.A.; Sato, H. The ketogenic diet upregulates expression of the gene encoding the key ketogenic enzyme mitochondrial 3-hydroxy-3-methylglutaryl-CoA synthase in rat brain. *Epilepsy Res.,* **2002**, *49*(2), 99-107. [http://dx.doi.org/10.1016/S0920-1211(02)00011-6] [PMID: 12049798]

[30] Fraser, D.D.; Whiting, S.; Andrew, R.D.; Macdonald, E.A.; Musa-Veloso, K.; Cunnane, S.C. Elevated polyunsaturated fatty acids in blood serum obtained from children on the ketogenic diet. *Neurology,* **2003**, *60*(6), 1026-1029. [http://dx.doi.org/10.1212/01.WNL.0000049974.74242.C6] [PMID: 12654976]

[31] Wexler, I.D.; Hemalatha, S.G.; McConnell, J.; Buist, N.R.; Dahl, H.H.; Berry, S.A.; Cederbaum, S.D.; Patel, M.S.; Kerr, D.S. Outcome of pyruvate dehydrogenase deficiency treated with ketogenic diets. Studies in patients with identical mutations. *Neurology,* **1997**, *49*(6), 1655-1661. [http://dx.doi.org/10.1212/WNL.49.6.1655] [PMID: 9409363]

[32] Lionel, A.C.; Monfared, N.; Scherer, S.W.; Marshall, C.R.; Mercimek-Mahmutoglu, S. MED23-associated refractory epilepsy successfully treated with the ketogenic diet. *Am. J. Med. Genet. A.,* **2016**, *170*(9), 2421-2425. [http://dx.doi.org/10.1002/ajmg.a.37802] [PMID: 27311965]

[33] Bough, K.J.; Gudi, K.; Han, F.T.; Rathod, A.H.; Eagles, D.A. An anticonvulsant profile of the ketogenic diet in the rat. *Epilepsy Res.,* **2002**, *50*(3), 313-325. [http://dx.doi.org/10.1016/S0920-1211(02)00086-4] [PMID: 12200222]

[34] Erecińska, M.; Nelson, D.; Daikhin, Y.; Yudkoff, M. Regulation of GABA level in rat brain synaptosomes: fluxes through enzymes of the GABA shunt and effects of glutamate, calcium, and ketone bodies. *J. Neurochem.,* **1996**, *67*(6), 2325-2334. [http://dx.doi.org/10.1046/j.1471-4159.1996.67062325.x] [PMID: 8931464]

[35] Wang, Z.J.; Bergqvist, C.; Hunter, J.V.; Jin, D.; Wang, D.J.; Wehrli, S.; Zimmerman, R.A. *In vivo* measurement of brain metabolites using two-dimensional double-quantum MR spectroscopy--exploration of GABA levels in a ketogenic diet. *Magn. Reson. Med.,* **2003**, *49*(4), 615-619. [http://dx.doi.org/10.1002/mrm.10429] [PMID: 12652530]

[36] Dahlin, M.; Elfving, A.; Ungerstedt, U.; Amark, P. The ketogenic diet influences the levels of excitatory and inhibitory amino acids in the CSF in children with refractory epilepsy. *Epilepsy Res.,* **2005**, *64*(3), 115-125. [http://dx.doi.org/10.1016/j.eplepsyres.2005.03.008] [PMID: 15961283]

[37] Yudkoff, M.; Daikhin, Y.; Horyn, O.; Nissim, I.; Nissim, I. Ketosis and brain handling of glutamate, glutamine, and GABA. *Epilepsia,* **2008**, *49*(Suppl. 8), 73-75. [http://dx.doi.org/10.1111/j.1528-1167.2008.01841.x] [PMID: 19049594]

[38] Al-Mudallal, A.S.; LaManna, J.C.; Lust, W.D.; Harik, S.I. Diet-induced ketosis does not cause cerebral acidosis. *Epilepsia,* **1996**, *37*(3), 258-261. [http://dx.doi.org/10.1111/j.1528-1157.1996.tb00022.x] [PMID: 8598184]

[39] Szot, P.; Weinshenker, D.; Rho, J.M.; Storey, T.W.; Schwartzkroin, P.A. Norepinephrine is required for the anticonvulsant effect of the ketogenic diet. *Brain Res. Dev. Brain Res.,* **2001**, *129*(2), 211-214. [http://dx.doi.org/10.1016/S0165-3806(01)00213-9] [PMID: 11506865]

[40] Masino, S.A.; Li, T.; Theofilas, P.; Sandau, U.S.; Ruskin, D.N.; Fredholm, B.B.; Geiger, J.D.; Aronica, E.; Boison, D. A ketogenic diet suppresses seizures in mice through adenosine A_1 receptors. *J. Clin. Invest.,* **2011**, *121*(7), 2679-2683. [http://dx.doi.org/10.1172/JCI57813] [PMID: 21701065]

[41] Juge, N.; Gray, J.A.; Omote, H.; Miyaji, T.; Inoue, T.; Hara, C.; Uneyama, H.; Edwards, R.H.; Nicoll, R.A.; Moriyama, Y. Metabolic control of vesicular glutamate transport and release. *Neuron,* **2010**, *68*(1), 99-112. [http://dx.doi.org/10.1016/j.neuron.2010.09.002] [PMID: 20920794]

[42] Kawamura, M., Jr.; Ruskin, D.N.; Masino, S.A. Metabolic autocrine regulation of neurons involves cooperation among pannexin hemichannels, adenosine receptors, and KATP channels. *J. Neurosci.,* **2010**, *30*(11), 3886-3895. [http://dx.doi.org/10.1523/JNEUROSCI.0055-10.2010] [PMID: 20237259]

[43] Tanner, G.R.; Lutas, A.; Martínez-François, J.R.; Yellen, G. Single K ATP channel opening in response to action potential firing in mouse dentate granule neurons. *J. Neurosci.,* **2011**, *31*(23), 8689-8696. [http://dx.doi.org/10.1523/JNEUROSCI.5951-10.2011] [PMID: 21653873]

[44] Manjarrez-Marmolejo, J.; Franco-Pérez, J. Gap junction blockers: An overview of their effects on induced seizures in animal models. *Curr. Neuropharmacol.,* **2016**, *14*(7), 759-771. [http://dx.doi.org/10.2174/1570159X14666160603115942] [PMID: 27262601]

[45] Maalouf, M.; Sullivan, P.G.; Davis, L.; Kim, D.Y.; Rho, J.M. Ketones inhibit mitochondrial production of reactive oxygen species production following glutamate excitotoxicity by increasing NADH oxidation. *Neuroscience,* **2007**, *145*(1), 256-264. [http://dx.doi.org/10.1016/j.neuroscience.2006.11.065] [PMID: 17240074]

[46] Milder, J.B.; Liang, L.P.; Patel, M. Acute oxidative stress and systemic Nrf2 activation by the ketogenic diet. *Neurobiol. Dis.,* **2010**, *40*(1), 238-244. [http://dx.doi.org/10.1016/j.nbd.2010.05.030] [PMID: 20594978]

[47] Kim, D.Y.; Davis, L.M.; Sullivan, P.G.; Maalouf, M.; Simeone, T.A.; van Brederode, J.; Rho, J.M. Ketone bodies are protective against oxidative stress in neocortical neurons. *J. Neurochem.,* **2007**, *101*(5), 1316-1326. [http://dx.doi.org/10.1111/j.1471-4159.2007.04483.x] [PMID: 17403035]