DIPONEGORO MEDICAL JOURNAL
(Jurnal Kedokteran Diponegoro)
Online : http://ejournal3.undip.ac.id/index.php/medico
E-ISSN : 2540-8844
Volume 11, Number 1, January 2022

THE CORRELATION OF LONG-TERM COMBINATION TREATMENT OF VALPROIC ACID AND TOPIRAMATE ON SERUM VITAMIN D LEVELS AMONG CHILDREN WITH EPILEPSY

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ABSTRACT

Background: Epilepsy is a chronic condition characterized by unprovoked seizure. Vitamin D is necessary in child development process and is strongly influenced by the presence of anti-epileptic drugs. **Objective:** To determine the correlation of treatment duration and serum vitamin D levels in children with epilepsy who use the combination of valproic acid and topiramate. **Methods:** A cross sectional study among forty pediatric patients with epilepsy with age range of 5-10 years at Dr. Kariadi Hospital Semarang. Examination of serum vitamin D levels is done by ELISA. Statistical analysis using Spearman correlation test and partial correlation test with significance level p = 0.05. **Results:** The mean of serum vitamin D levels in patient with combination treatment after 24 months was 21.87±1.21ng/ml, in those who still having seizure was 27.94±2.40ng/ml, and among who received delayed therapy was 23.23±2.07ng/ml. Bivariate analysis depicted that there was negative correlation between duration of treatment (rho= -0.850, p<0.001), onset of therapy (rho=0.604, p<0.001) and seizure frequency (rho=0.559, p<0.001) with serum vitamin D levels. Partial correlation analysis showed a significant correlation between duration of treatment and serum vitamin D levels after adjusted by onset of therapy (rho=-0.839, p<0.001) and seizure frequency (rho=-0.856, p<0.001). **Conclusion:** There is a correlation between the duration of treatment and serum vitamin D levels among children with epilepsy

Keywords: *Epilepsy, Valproic Acid, Vitamin D, Topiramate*

INTRODUCTION

Epilepsy is a common neurological disease that occurs at all ages. It is defined as a condition that is prone to recurrent seizure, at least 2 or more unprovoked seizure after 24 hours. Epilepsy is commonly occurred in children with the consequence of the long-term antiepileptic drug therapy. Several studies have reported that the incidence of epilepsy in the first year of life is about 100 cases per 100,000 children.[1]

Anti-epileptic drugs are used to prevent and stop recurrent seizure. Seizure that lasts in long time will cause damage to the death of neuron cells. The principle in administering antiepileptic drug therapy begins with monotherapy according to the type of seizure and epilepsy syndrome. Some antiepileptic drugs induce P-450 enzyme in the liver which may cause vitamin D deficiency. However, some drugs that do not induce this enzyme may also cause vitamin D hypovitaminosis.[2,3]

Vitamin D is a one of the steroid hormone family which is stimulated through nuclear and membrane-related receptors.[4] It is important in the process of child development, especially in bone development and growth. Deficiency of vitamin D in children is most often caused by the use of antiepileptic drugs in long term, where it will cause a decrease in bone mineral density thereby increasing the risk of fracture.[5] Vitamin D is also an important component for the development of brain function and growth, especially in children.[6]

Dr. Kariadi National Hospital Semarang is the largest hospital as well as a referral hospital for the Central Java region. Currently Dr. Kariadi National Hospital is a class A teaching hospital and functions as a teaching hospital for doctors, specialists, and sub-specialists from the Faculty of Medicine, Diponegoro University. This hospital serves National Health insurance. Patients who are treated at this hospital, especially epilepsy, received combination therapy with anti-epileptic drugs including a combination of valproic acid and topiramate, which is the most often combination given to paediatrics patients with epilepsy.

METHOD

This study is a cross-sectional study, which was conducted at the outpatient Department of Neurology, Dr Kariadi National Hospital, Semarang from November 2020 to January 2021. The subjects

were pediatric epilepsy patients who received a combination of valproic acid and topiramate after 6 months, 12 months, and 24 months with an age range of 5-10 years. Meanwhile, the exclusion criteria for pediatric patients were obese or undernourished, impaired kidney function, patients taking vitamin D supplementation, and severe side effects due to valproic acid and topiramate therapy.

This research has received permission from the Ethics Committee of the Faculty of Medicine Diponegoro University with the number 619/EC/KEPK-RSDK/2020 dated September 14, 2020. Patients were selected based on inclusion and exclusion criteria, then informed consent was given to the patient. The study began with history and physical examination. After that a blood sample was taken to check vitamin D levels. After completion, the blood sample sent to the GAKI Laboratory.

The data were processed using SPSS Statistics for Windows version 23 program to determine the correlation between the duration of treatment with the combination of valproic acid and topiramate on serum vitamin D levels. Correlation test was conducted with a 95% confidence level. The confounding variables were the frequency of seizures, initiation of therapy and age of epilepsy onset. These variables were tested using a bivariate test with the Spearman correlation test. Gender was tested using chi-square test. Then a partial correlation test will be carried out by controlling the frequency of seizures and starting therapy with the significance level p = 0.05.

RESULTS

From the demographic data in table 1, there were more male patients than female patients. The most subjects were received the combination treatment of valproic acid and topiramate after 24 months (42.5%). The age of seizure onset at most was <1 year (40%). The study also showed more patient with seizure (65%) and patient who received early therapy (60%).the average vitamin D level was 33.11 ng/ml.

Table 1. Demographic and Clinical Data

Variable	F	%	Mean ± SD	Median (min – max)
Demographic Data				
Age			8.55 ± 1.89	10 (5-10)
Gender				
Male	26	65.0		
Female	14	35.0		
Clinical Data				
Duration of Treatment				
After 6 months	12	30.0		
After 12 months	11	27.5		
After 24 months	17	42.5		
Age of seizure onset				
< 1 years old	16	40.0		
1-5 years old	10	25.0		
> 5 years old	14	35.0		
Frequency of Seizure				
Seizure	26	65		
No Seizure	14	35		
Initiation of Therapy				
Early Therapy	24	60	8.13 ± 9.258	
Late Therapy	16	40		
Mean of Vitamin D Serum Level			33.11 ± 13.73	31.15 (12.10 – 68.20)

Table. 2 Mean Serum Vitamin D Levels based on Duration of Treatment, Frequency of Seizures, Initiation of Therapy, and Age of Seizure Onset

Variable	Vitamin D serum level (Mean (ng/ml))
Duration of Treatment:	
After 6 months	49.79±2.62
After 12 months	32.31±2.24
After 24 months	21.87±1.21
Frequency of Seizure	
No Seizure	42.73±2.98
Seizure	27.94±2.40
Initiation of Therapy	
Early Therapy	39.71±2.59
Late Therapy	23.23±2.07
Age of seizure onset	
< 1 years old	34.26±3.65
1-5 years old	31.76±3.84
>5 years old	32.77±3.92
Gender	
Male	33.13±2.69
Female	33.07±3.86

Table 2shows the average vitamin D level to the duration of combination drug valproic acid and topiramate. which is the longer treatment will further reduce serum vitamin D levels. It is also assumed that patients who are still having seizure will show lower serum vitamin D levels. In addition, patients with epilepsy who are delayed in receiving anti-epileptic drug therapy will have lower serum vitamin D levels. This study also showed that the age of seizure onset did not affect serum vitamin D levels. The mean serum vitamin D levels in men and women are relatively the same.

Table 3. The Correlation of Gender to Serum Vitamin D Levels

Variable	Vit. D		p
	Normal	Abnormal	
Gender			
Male	13 (50%)	13 (50%)	0,666
Female	8 (57,1%)	6 (42,9%)	

* Chi-square correlation test, significant if p<0.05

In this study, because it uses a nominal and ordinal scale with an abnormal distribution, the Chi-square test can be performed. As a result, there was no significant difference between sex and serum vitamin D levels (p=0.666).

Table 4. The Correlation between duration of treatment, frequency of seizures, initiation of therapy, and age of seizure onset on serum vitamin D levels

Variable	Rho	P
Duration of Treatment	-0.850	0.001*
Frequency of Seizure	0.559	0.001*
Initiation of Therapy	0.604	0.001*
Age of seizure onset	0.044	0.786

* Spearman Correlation Test, significant if p<0.05

In the study of the correlation between duration of treatment, frequency of seizures, starting therapy, and age of seizure onset on serum vitamin D levels, the data distribution was not normal, so the analysis was carried out using the Spearman correlation test. This study resulted in a statistically significant correlation between duration of treatment, frequency of seizures, and initiation of therapy on serum vitamin D levels (p=0.001). However, there was no statistically significant correlation between age of seizure onset and serum vitamin D levels (p=0.786).

Table 5. The Correlation between Valproic acid and Topiramate Combination Treatment Duration of Vitamin D Serum Levels by Controlling Seizure Frequency and Starting Therapy

Variable	Controlling Variable	rho	p
Vitamin D	Initiation of Therapy	-0.839	0.001*
	Frequency of seizure	-0.856	0.001*

* Significant Partial Correlation Test if p<0.05

In this study, we correlated the duration of valproic acid and topiramate combination treatment with serum vitamin D levels controlled by initiation of therapy, resulting in a strong strength result (rho=-0.839) which was statistically significant (p<0.001). Meanwhile, the correlation between duration of valproic acid and topiramate combination treatment with serum vitamin D levels controlled by frequency of seizures resulted in a strong correlation strength (rho=-0.856) which was statistically significant (p<0.001).

DISCUSSION

In this study more male subjects than female, this is in accordance with the general epidemiology where the male sex is slightly more than female.[7]

Choong Yi Fong and Catherine Riney who conducted research on epileptic children in South Queensland, Australia with 111 research subjects revealed that men and women had no effect on serum Vitamin D levels (p = 0.696), which was also less more the same as this study (p=0.666).[8] This statement is also supported by Saket et al. with 60 participants and 60 controls, that showed serum vitamin D levels had no significant difference between gender (p=0.875).[9]

We found that the longer duration of valproic acid and topiramate combination treatment, the lower the serum vitamin D level (mean serum vitamin D levels after 6 months of treatment is 49.79 ng/ml, after 12 months 32.31ng/ml, and after 24 months 21.87ng /ml). This is in accordance with the research conducted by Yun Jin Lee et al. where a study in Yangsan, Korea with subject of 143 pediatric epilepsy patients revealed that treatment carried out for 2 years resulted in low serum vitamin D levels ($p < 0.01$) and the same statement in a study conducted by Choong Yi Fong and Catherine Riney ($p<0.01$).[8,10] A study from Indonesia conducted by Narulita Laksmi et al. at Pediatric Neurology Department of Moewardi Hospital Solo stated that the duration of antiepileptic drug therapy had a significant difference in serum Vitamin D levels (p=0.03).[11] This is in accordance with the theory that antiepileptic drugs have the effect of lowering serum Vitamin D levels, especially through induction of cytochromes P-450 which is an enzyme that helps catabolism of Vitamin D.[12]

Research by Holo et al. with 14 patients who had seizure and received vitamin D supplementation, showed 40% reduction in seizure. It consistent with our assumption that convulsion that occur continuously will cause a decrease in serum vitamin D levels (the average serum vitamin D level for patients who do not have seizures is 42.71ng/ml and those who are still having seizures is 27.94ng/ml). Vitamin D has anti-inflammatory activity that contributes to preventing the inflammatory activity of IL-1 and IL-6 which occurs in the inflammatory process of epilepsy.[13]

Our study also assumed that the longer patient receives anti-epileptic drug therapy from the first seizure, the lower the serum vitamin D level (the mean serum vitamin D level in patients who received early therapy was 39.71ng/ml and those who received delayed therapy 23.23ng/ml). It is supported by the theory put forward by Nikoles E. Koundourakis and Andrew N. Margioris which revealed that vitamin D receptors are abundant along neurons and glial cells, where vitamin D receptors function is activating the passive form of vitamin D to become active with the help of enzymes. A precursor of vitamin D activator (1-α hydroxylase) is also produced by brain cells. Seizure that occurs continuously will cause neuronal damage due to the inflammatory process that causes the process of apoptosis and nerve necrosis.[14]

This study showed mean serum vitamin D levels at the age of seizure onset were approximately the same. This is in accordance with the research conducted by Chong Yi Fong and Catherine Riney where the age of onset did not have a significant difference ($p = 0.855$).[8]

On the analysis of the correlation between duration of treatment with valproic acid and topiramate combination and serum vitamin D levels showed statistically significant strong correlation with negative correlation direction (rho=-850; $p<0.001$). This is in accordance with the research conducted by Yun Jin Lee et al. where the study in children with epilepsy revealed that treatment given for 2 years resulted in lower serum vitamin D levels ($p<0.01$). The same statement was also expressed by Choong Yi Fong and Catherine Riney ($p<0.01$).[8,15] This is in accordance with the theory where there are 2 groups of anti-epileptic drugs, those that induce liver enzymes and those that not. Antiepileptic drugs that induce liver enzymes cause the release of cytochrome P-450 enzyme, where functions in the catabolism of vitamin D and reduces the calcium absorption process so that it will have a long-term impact on bone mineral content in the child's growth process. Meanwhile, drugs that do not induce liver enzymes have effect on stimulating the steroid hormone xenobiotic receptor which induces the 25-hydroxyvitamin D-24-hydroxylase enzyme. This enzyme functions as a catabolism of vitamin D.[16,17,18] Antiepileptic drugs will increase the risk of developing low levels of Vitamin D. This is evidenced by several studies which state polytherapy has more significant differences in the occurrence of low vitamin D levels compared to

monotherapy treatment, one of which is the study by Yun Jin Lee and et al. ($p<0.01$).[15]

Our analysis on the correlation between seizure frequency and serum vitamin D levels, there was a statistically significant moderate strength correlation with a positive correlation direction (rho=0.559; $p<0.001$). This agrees with several studies which state that seizure can occur due to low levels of vitamin D, especially in several studies that are associated with patients with hereditary or nutritional rickets.[19] The first research on 1974 proposed that vitamin D supplement may reduce mean seizure frequency by up to 30% after therapy. In another study conducted by Hollo et al. (2012) from 14 patients who experienced seizures with Vitamin D supplementation, there was an increase in seizure followed by a significant decrease in seizure frequency ($p = 0.04$) with an average decrease of seizure by 40% and 5 study subjects who experienced a reduction in seizures 50%.[20] This is in accordance with the theory expressed by Harms Lauren et al. that vitamin D supplementation increase levels of antioxidants, such as glutathione in the brain. In addition, vitamin D can also inhibit neuronal uptake of ROS such as hydrogen peroxidase and protect the excitability of glutamate so that the protection against excitotoxicity provided by vitamin D can also protect against seizure It has been proven in rat animal models where vitamin D increase the electroconvulsive seizure threshold and reduce seizure severity.[21]

An analysis of the correlation between initiation therapy and serum vitamin D levels, showed strong statistically significant strength with a positive correlation direction (rho=0.604; $p<0.001$). This is in accordance with the theory that vitamin D levels can be affected by neuron brain damage. In the process of seizure, there are many metabolic changes that cause neuronal damage, especially hypoxia, lactic acidosis, CO_2 necrosis, hyperkalemia, hypoglycemia which can cause brain neuron damage.[22] N. Margioris revealed that vitamin D receptors are abundant along brain neuron cells including in the primary motor cortex, especially in neurons and glial cells in several brain areas including cortex, deep gray matter, cerebellum, brain stem nucleus, spinal cord and ventricular system. In addition, 1-α-hydroxylase enzyme, a precursor of vitamin D activator (), is also produced by the brain. Vitamin D levels are also associated with conduction velocity by motor neurons mediated by the neuro transmitters dopamine, serotonin, acetylcholine, GABA, and catecholamines. They stated that vitamin D levels are inversely related to oxidative stress which cause brain damage by apoptosis and neuronal necrosis.[14]

An analysis of the correlation between duration of valproic acid and topiramate treatment combination and serum vitamin D levels controlled by initiation therapy, showed statistically significant result. Therefore after initiation therapy was controlled, the strength of the correlation was still strong, which means this correlation is pure without bias by starting therapy. In addition, the analysis of the correlation between duration of treatment with a combination of valproic acid and topiramate which was controlled by the frequency of seizures, showed that the strength of the correlation was statistically significant. Hence after the seizure frequency was controlled, the strength of the correlation was still strong, which means that this correlation was purely without bias by the frequency of seizure. Theoretically both of these are in accordance with the theory that antiepileptic drugs have the effect of lowering vitamin D levels, mainly through the induction of cytochrome P-450. This mechanism is also mediated by the pregnane X-receptor (PXR). Activation of PXR by antiepileptic drugs results in the induction of CYP2 and CYP3, which are cytochrome enzymes involved in drug metabolism. PXR activators such as antiepileptic drugs can increase CYP24 expression. CYP24 is a major enzyme that affects the metabolism of 1.25(OH)2D and thereby reduces the active form of Vitamin D.[23-25]

Our study has limitations. First, we lack comprehensive dietary assessment and home backgrounds in our patients. Second, we did not monitor the patient's sun exposure which could affect vitamin D levels.

CONCLUSION

We can conclude that there was a strong correlation between duration of treatment with combination of valproic acid and topiramate and serum vitamin D levels in children with epilepsy. There were a strong and moderate correlation between vitamin D serum with initiation of therapy and frequency of seizure.

Ethical Clearance

The research permission from the Ethics Committee of the Kariadi Hospital Semarang with the number 619/EC/KEPK-RSDK/2020 dated September 14, 2020.

Conflicts of Interest

The authors declare no conflict of interest.

Funding

This research did not receive any specific grant from funding agencies in the public, commercial, or not for profit sector.

Author Contributions

For research articles with several authors, a short paragraph specifying their individual contributions must be provided. The following statements should be used as follows: "conceptualization, Hexanto Muhartomo and MI Widiastuti Samekto; methodology, MI Widiastuti Samekto; software, Rony Parlindungan Sinaga; validation, Endang Kustiowati, Aris Catur Bintoro, and Arinta Puspita Wati; formal analysis, Hexanto Muhartomo; investigation, Hexanto Muhartomo; resources, Alifiani Hikmah Putranti; data curation, Hexanto Muhartomo; writing—original draft preparation, Hexanto Muhartomo; writing—review and editing, Rony Parlindungan Sinaga; visualization, Hexanto Muhartomo; supervision, MI Widiastuti; project administration, Rony Parlindungan Sinaga; funding acquisition, Hexanto Muhartomo

Acknowledgments

We would like to thank Neurology and Pediatric Neurology Department at dr. Kariadi Central General Hospital Semarang who supported this study, Neurology Lecturer of Medical Faculty Diponegoro University, for their guidance and assistance in this study. The authors state that they had no interests that could be perceived as posing a conflict or bias.

REFERENCES

1. Saad K. Childhood Epilepsy : An Update on Diagnosis and Management Childhood. 2015;(January).
2. Kusumastuti K, Gunadharma S, Kustiowati E. PEDOMAN TATALAKSANA EPILEPSI. Surabaya: Airlangga University Press; 2019. p. 1.
3. Kwan P, Brodie MJ. Combination Therapy in Epilepsy. Drugs. 2006;66(14):1817–29.
4. Holló A, Clemens Z, Kamondi A, Lakatos P, Szucs A. Correction of vitamin D deficiency improves seizure control in epilepsy: A pilot study. Epilepsy Behav. 2012;24(1):131–3.
5. Abdullah AT, Mousheer ZT. Vitamin D Status in Epileptic Children on Valproic acid; a Case-Control Study. Arch Acad Emerg Med. 2020;8(1):e13.
6. Chowdhury R, Taneja S, Kvestad I, Hysing M, Bhandari N, Strand TA. Vitamin D status in early childhood is not associated with cognitive development and linear growth at 6-9 years of age in North Indian children: A cohort study. Nutr J. 2020;19(1):1–9.
7. Minardi C, Minacapelli R, Valastro P, Vasile F, Pitino S, Pavone P, et al. Epilepsy in children: From diagnosis to treatment with focus on emergency. J Clin Med. 2019;8(1):1–10.
8. Fong CY, Riney CJ. Vitamin D deficiency among children with epilepsy in South Queensland. J Child Neurol. 2014;29(3):368–73.
9. Saket S, Varasteh N, Halimi Asl AA, Saneifard H. How antiepileptics may change the serum level of vitamin d, calcium, and phosphorus in children with epilepsy. Iran J Child Neurol. 2021;15(1):19–27.
10. Lee JY, So T-Y, Thackray J. A Review on Vitamin D Deficiency Treatment in Pediatric Patients. J Pediatr Pharmacol Ther. 2013;18(4):277–91.
11. Tantri NL, Nur FT, Salimo H. Pengaruh Pemberian Obat Antiepilepsi terhadap Kadar Vitamin D pada Anak Penderita Epilepsi. Sari Pediatr. 2017;19(2):97.
12. Zhou C, Assem M, Tay JC, Watkins PB, Blumberg B, Schuetz EG, et al. Steroid and xenobiotic receptor and vitamin D receptor crosstalk mediates CYP24 expression and drug-induced osteomalacia. 2006;116(6):1703–12.
13. Holick MF. Vitamin D and brain health: The need for vitamin D supplementation and sensible sun exposure. J Intern Med. 2015;277(1):90–3.
14. -Galvan, R. F, Barranco V, Galvan JC, Batlle, Sebastian FeliuFajardo S, García. Vitamin D and Physical Activity. Intech [Internet]. 2016;i(tourism):13.
15. Lee YJ, Park KM, Kim YM, Yeon GM, Nam

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(Jurnal Kedokteran Diponegoro)
Online : http://ejournal3.undip.ac.id/index.php/medico
E-ISSN : 2540-8844
Volume 11, Number 1, January 2022

SO. Longitudinal change of vitamin D status in children with epilepsy on antiepileptic drugs: Prevalence and risk factors. Pediatr Neurol. 2015;52(2):153–9.
16. Menon B, Harinarayan C V. The effect of anti epileptic drug therapy on serum 25-hydroxyvitamin D and parameters of calcium and bone metabolism-A longitudinal study. Seizure. 2010;19(3):153–8.
17. Purniti PS. Paediatrica Indonesiana. 2011;51(4):207–12.
18. Eyles DW, Burne THJ, McGrath JJ. Vitamin D, effects on brain development, adult brain function and the links between low levels of vitamin D and neuropsychiatric disease. Front Neuroendocrinol. 2013;34(1):47–64.
19. Kalueff A V., Minasyan A, Keisala T, Kuuslahti M, Miettinen S, Tuohimaa P. Increased severity of chemically induced seizures in mice with partially deleted Vitamin D receptor gene. Neurosci Lett. 2006;394(1):69–73.
20. Durá-Travé T, Gallinas-Victoriano F, Malumbres-Chacón M, Moreno-Gónzalez P, Aguilera-Albesa S, Yoldi-Petri ME. Vitamin D deficiency in children with epilepsy taking valproate and levetiracetam as monotherapy. Epilepsy Res. 2018;139(November 2017):80–4.
21. Harms LR, Burne THJ, Eyles DW, McGrath JJ. Vitamin D and the brain. Best Pract Res Clin Endocrinol Metab. 2011;25(4):657–69.
22. Wasterlain CG, Fujikawa DG, Penix LR, Sankar R. Pathophysiological Mechanisms of Brain Damage from Status Epilepticus. Epilepsia. 1993;34(April 2018):S37–53.
23. T. Chen, Z. Lu MFH. Photobiology of Vitamin D, in: M.F. Holick (Ed.), Vitamin D. Vol. 25, Molecular Biology and Clinical Application. 2010. 175–187 p.
24. Wang Z, Lin YS, Zheng XE, Senn T, Hashizume T, Scian M, et al. An inducible cytochrome P450 3A4-dependent vitamin D catabolic pathway. Mol Pharmacol. 2012;81(4):498–509.
25. Benedetti MS. Enzyme induction and inhibition by new antiepileptic drugs : a review of human studies. 2000;14:301–19.