Comparison of the Serum Concentrations of Micronutrients Zinc and Iron in Epileptic Children before and after Administration of Valproic ACID

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Abstract

Introduction: The mortality rate of epileptic patients has increased despite advances in the treatment of epilepsy. Several studies have indicated that changes of micronutrient homeostasis led to seizures. The anticonvulsant valproic acid affects the intracellular concentration of micronutrients.

Objective: To compare the serum concentrations of micronutrients zinc and iron in paediatric patients with epilepsy before and after administration of valproic acid.

Method: A prospective cohort design study was conducted in children with epilepsy from July to December 2019 in the outpatient department of paediatric neurology, Dr. Soetomo Regional Public Hospital, Surabaya, Indonesia. Serum concentrations of micronutrients zinc and iron were measured using the colorimetric method. Statistical analysis used paired t-test with $p<0.05$ being considered significant.

Results: The study sample comprised 20 children with epilepsy. There were 13 (65%) males. Eight (40%) children were in the 1-5 year age group. Seizure type was generalised in 18 (90%) patients. The mean serum iron concentrations before and after valproic acid treatment were 23.9±16.8 µmol/L and 25.8±16.4 µmol/L respectively ($p=0.700$). The mean serum zinc concentrations before and after valproic acid treatment were 13±16.8 µmol/L and 26.8±15.4 µmol/L respectively ($p=0.004$).

Conclusions: There was a statistically significant increase in the mean serum zinc concentration after valproic acid treatment in children with epilepsy. There was however no statistically significant increase in the mean serum iron concentration after valproic acid treatment in children with epilepsy.

Key words*: epilepsy, micronutrients iron and zinc, valproic acid, children.*

Introduction

Globally, 50 million people are affected by epilepsy with more than half of the cases occurring in children[1]. In Indonesia, there are 700.000-1.400.000 cases of epilepsy, with 70.000 new cases every year and about 40-50% of these are children[2]. Valproic acid affects intracellular zinc concentration, the zinc transporter (ZnT1) playing a role in intracellular zinc homeostasis. Serum zinc concentration may decrease as a result of valproic acid, being affected by the dietary intake of zinc, stress, serum albumin concentration and the metabolic state of the patient[3]. An alteration of iron metabolism, consisting of an increase of non-transferrin bound iron (NTBI), occurred in 24 epileptic patients receiving valproic acid monotherapy[4]. This study was conducted

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to observe the effect of anti-epileptic drug valproic acid on the serum concentrations of zinc and iron.

Objectives

To compare the serum concentrations of micronutrients zinc and iron in paediatric patients with epilepsy pre- and post-administration of valproic acid.

Method

A prospective cohort observational analytical study was conducted from July to December 2019 in children aged 18 years or less with newly diagnosed epilepsy in the Outpatient Department of Paediatric Neurology, Dr. Soetomo Regional Public Hospital, Surabaya, Indonesia, using valproic acid monotherapy. The characteristics of the study population (age, sex, seizure type, laboratory examination and electroencephalogram (EEG) were recorded. Serum concentrations of micronutrients iron and zinc were measured using the colorimetric method. Statistical analysis used paired t-test and $p<0.05$ was considered significant.

A written statement of agreement was obtained from the parents or guardians for the child's participation in the study. Ethics Approval was obtained from Health Research Ethical Committee of Regional Public Hospital of Dr. Soetomo Surabaya before commencing the study.

Results

The study sample comprised 20 children. The basic characteristics of the study sample are shown in Table 1.

Table 1: ***Basic characteristics of study sample (n=20)***

Variable	Result
Sex - n (%)	
Male	13 (65)
Female	07 (35)
Age in years – mean ± SD	4.2 ± 4.23
< 1 year - n (%)	05 (25)
1-5 years - n (%)	08 (40)
>5 years - n (%)	07 (35)
Nutritional status - n (%)	
Normal	13 (65)
Malnutrition	07 (35)
Type of seizure - n (%)	
General	18 (90)
Partial	02 (10)
Family history of epilepsy - n (%)	
Yes	05 (25)
No	15 (75)
Haemoglobin (g/dl), mean ±SD	11.4 ± 1.29
Normal	10 (50)
Anaemia	10 (50)
Electroencephalogram (EEG) - n (%)	
Normal	11 (55)
Abnormal	09 (45)

Of the 9 cases with abnormal electroencephalograms (EEGs), 3 had benign rolandic epilepsy.

Serum iron and zinc levels before and after valproic acid therapy are shown in Table 2.

Table 2: ***Serum iron and zinc levels before and after valproic acid therapy***

Laboratory	Before		After	
	n (%)	Mean ± SD	n (%)	Mean ± SD
Serum iron levels High (≥24 µmol/L) Low (≤8 µmol/L) Normal (8-24 µmol/L)	 10 (50) 06 (30) 04 (20)	23.9 ±16.8	 07 (35) 01 (05) 12 (60)	25.8 ±16.4
Serum zinc levels High (≥20 µmol/L) Low (≤11 µmol/L) Normal (11-20 µmol/L)	 04 (20) 10 (50) 06 (30)	13 ±16.8	 14 (70) 0 06 (30)	26.8 ±15.4

The mean serum iron concentration before treatment was 23.9 ± 16.8 µmol/L and the mean serum zinc concentration before treatment was 13 ±16.8 µmol/L. The mean serum iron concentration after valproic acid treatment was 25.8 ±16.4 µmol/L and the mean serum zinc concentration after valproic acid treatment was 26.8 ±15.4 µmol/L. The results of the analysis using paired t-test showed a significant difference between the mean serum concentrations of zinc before and after valproic acid treatment in patients with epilepsy ($p = 0.004$). However, there was no significant difference found between the mean serum concentrations of iron before and after valproic acid treatment in patients with epilepsy ($p = 0.700$).

The correlation between anaemia and serum iron and zinc levels after valproic acid therapy is shown in Table 3.

Table 3: ***Correlation between anemia and iron and zinc levels after being treated with valproic acid***

Laboratory	Serum iron levels after valproate acid therapy			p*	Serum zinc levels after valproate acid therapy			p*
	Low	High	Normal		Low	High	Normal	
Haemoglobin Anaemia Normal	 1 0	 7 0	 10 2	0.558	 0 0	 12 2	 6 0	1.000

**p < 0.05 was considered statistically significant; Chi Square test was used*

Correlation of serum iron and zinc levels before and after valproate acid therapy is shown in Table 4.

Table 4: ***Correlation of serum iron and zinc levels before and after valproic acid therapy***

Variable	Before and after valproate acid therapy	p value
Serum iron levels (mean ± SD)	1.90 ± 21.8)	0.700
Serum iron levels (mean ± SD)	13.79 ± 18.6)	0.004*

**p <0.05 was considered statistically significant*

Discussion

In this study of epileptic children 65% were males. This was similar to the study by Serdaroglu *et al* (2004) where there was male predominance[5]. In our study 40% were in the 1-5 year age group. Topbas *et al* (2012) also found most epileptics to be 1-5 years old[6]. In our study 50% of patients were anaemic. Zareifar *et al* (2012), reported that haemoglobin concentration was significantly lower in children with epilepsy[7]. In our study only 25% of patients had a family history of epilepsy. This is similar to the study by Omar *et al* (2016) where there was a family history of epilepsy in 25% cases[8]. In our study 90% had generalised seizures. In the study by Andrianti *et al* (2016) generalised seizures were the predominant type[9]. EEG examination is vital to confirm the clinical diagnosis of epilepsy, classification of epilepsy type, epileptogenic focus, treatment evaluation, and prognosis determination[10]. However, the first EEG was normal in 55% of our cases. However, it should be remembered that the EEG was done when the patient was not in seizure (inter-ictal). In our study 35% of children with epilepsy were malnourished. In a study by Hardaningsih *et al* (2016) malnutrition status based on WLZ score <-2SD proved to be one of status epilepticus risk factors in children with seizure[11].

The zinc status in man is measured by the zinc concentration in plasma and serum. However, this is not a good indicator since less than 1% of the zinc total in the body is circulating in the plasma and also because the plasma zinc concentration is affected by hypoalbuminemia, haemoconcentration and acute phase response. Children in developing countries received inadequate zinc intake[12]. A study by Soltani *et al* (2016), explained that zinc concentration decreased in children with epilepsy before starting administration of anti-epileptic drugs[13]. In our study, children with epilepsy, prior to receiving valproic acid treatment, had low serum zinc concentrations in 10 (50%), normal serum zinc concentrations in 6 (30%) and high serum zinc concentrations in 4 (20%). The mean zinc concentration was 13±16.8 µmol/L.

A study conducted in man about valproate acid mechanism in zinc metabolism remains controversial[14]. This study obtained a mean serum zinc concentration of 26.8 ±15.4 µmol/L after the administration of valproic acid. This indicated an increase of zinc concentration compared to that before the administration of valproic acid. High concentrations were obtained in 14 (70%) patients and normal concentrations in 6 (30%) patients. Two patients had the same high zinc serum concentrations before and after the administration of valproic acid. Analysis using paired t-test showed a significant difference of mean serum zinc concentration before and after valproic acid in patients with epilepsy ($p = 0.004$). Zinc serum concentrations might be higher as it took a longer time to obtain serum after blood uptake. During the blood clotting to obtain serum, zinc could be released from the blood cell and thrombocyte in such a way that zinc concentration in the serum increased.

There was an increase in the concentration of several micronutrients in groups receiving anti-epileptic treatment as a result of improved absorption of metal from the digestive tract or decrease elimination from the body[15]. This study also found normal zinc serum concentration, as a result of 1% zinc serum concentration from all zinc of the body and did not describe the state

of total zinc in the body[15]. According to Verrotti *et al* (2002), the effect of long term administration of valproic acid treatment in 36 patients with epilepsy before and after the 1 year-treatment showed that zinc concentration was normal[16].

In our study, children with epilepsy, before the administration of valproic acid had low serum iron concentrations in 6 (30%) and normal serum iron concentrations in 14 (70%) patients. The mean serum iron concentration in children with epilepsy before administration of valproic acid was 23.9 ± 16.8 µmol/L and mean serum iron concentration after administration of valproic acid was 25.8±16.4 µmol/L. This indicated an increase of iron serum concentration before and after the administration of valproic acid treatment. However, in this study, after the administration of valproic acid, serum iron concentration was normal in 12 patients, low in one patient and high in 7 patients. From the paired t-test, it was shown that there was no significant difference in mean serum iron concentrations before and after valproic acid treatment in patients with epilepsy (p = 0.700).

The serum iron level in each individual may vary 10-40% because of the change in iron absorption, bone marrow uptake on iron, or iron deposit expenditure. Besides, iron serum concentration is determined by iron absorption in the intestines, iron deposit, the speed of haemoglobin breakage and new haemoglobin formation[17].

The increase in serum iron concentration after valproic acid treatment resulted from change in iron homeostasis; consequently, the free iron in plasma or non-transferrin-bound (NTBI) iron began to accumulate in the blood and this resulted in increased serum iron concentration. The NTBI iron was finally inserted in the tissue by an unknown mechanism, which caused the impairment of cells and tissues. In most patients, excessive iron was a result of NTBI uptake from the circulation. Similar results were found in the study by Ounjaijean *et al* (2011), stating that 24 patients with epilepsy receiving VPA monotherapy caused an iron metabolism change, therefore an increase was found in NTBI[4]. In our study, mean serum iron concentration was normal after valproic acid treatment, similar to the study by Soltani *et al* (2016), where serum iron concentration did not change[13].

Conclusions

Serum concentration of iron and zinc on pediatric patients with epilepsy increased after valproate acid treatment.

Conflict of Interest : The authors declare that there is no conflict of interest regarding this research.

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Ethical Clearance: This study was approved by the Ethical Committee of Dr. Soetomo General Hospital, Surabaya No. 1547/KEPK/X/2019

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