

CHANGES IN HOMOCYSTEINE LEVELS IN EPILEPTIC CHILDREN RECEIVING SODIUM VALPROATE

Sodyum valproat alan epileptik çocuklarda homosistein düzeylerindeki değişiklikler

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Abstract

Purpose: The data regarding valproate and their influence on folate and homocysteine levels are conflicting. The aim of this study was to evaluate whether differences exist in homocysteine, folate, and vitamin B₁₂ levels in children receiving valproate.

Patients and Methods: Sixty-five patients receiving sodium valproate and twenty-five healthy controls were included in the study. Serum total homocysteine levels were analyzed by enzyme linked immunosorbent assay kit. Vitamin B₁₂ and folate values were measured by Bio DPC kits.

Results: Serum homocysteine concentrations were significantly higher in patients than those of the controls. Moreover, serum folate levels were observed to be significantly decreased in patients compared to controls. On the contrary, serum vitamin B₁₂ levels did not change in the patients.

Conclusion: Our data show that prolonged treatment with valproate increases serum homocysteine concentrations suggesting that an effect of inducer valproate on homocysteine metabolism could exist, which is not independent of the effect of low folate levels. These results indicate that homocysteine levels should be determined in order to identify hyperhomocysteinemia among patients receiving sodium valproate.

Key Words: Epilepsy, Folic acid, Homocysteine, Valproate

In recent years, plasma total homocysteine (tHcy) level and its association with cardiovascular diseases have received considerable attention. Vitamin B₁₂ and folate function as coenzymes in the metabolism of Hcy. It is suggested that deficiency of these vitamins is associated with increased plasma concentrations of Hcy (1, 2). Ono

Özet:

Amaç: Valproatın folat ve homosistein düzeyleri üzerinde etkileri ile ilgili veriler çelişkilidir. Bu çalışmanın amacı valproat alan çocuklarda homosistein, folat ve vitamin B₁₂ düzeylerinde farklılık olup olmadığını değerlendirmektir.

Hastalar ve Yöntem: Sodyum valproat alan altmışbeş hasta ve yirmibeş sağlıklı kontrol çalışıldı. Serum total homosistein düzeyleri ELİZA kitiyle analiz edildi. Vitamin B₁₂ and folat değerleri Bio DPC kitleriyle ölçüldü.

Bulgular: Serum Hcy konsantrasyonları hastalarda kontrollerdekinden anlamlı olarak daha yüksekti. Üstelik, kontrollerle karşılaştırıldığında serum folat düzeylerinin hastalarda anlamlı olarak azaldığı gözlemlendi. Bunun aksine, serum vitamin B₁₂ düzeyleri değişmedi.

Sonuç: Sonuçta, bizim verilerimiz valproat ile uzun süre tedavinin serum homosistein konsantrasyonlarını arttırdığını göstermektedir; homosistein metabolizması üzerinde valproatın indükleyici bir etkisi olabilir ve bu etki düşük folat düzeylerinden bağımsız değildir. Bu sonuçlar sodyum valproat alan hastalarda hiperhomosisteineminin tanımlanması için homosistein düzeylerinin tayin edilmesi gerektiğini göstermektedir.

Anahtar Kelimeler: Epilepsi, Folik asit homosistein, Valproat

reported that 4 of 130 epileptic patients receiving various AEDs had low plasma folate and elevated Hcy concentrations (3). Schwanninger found that mean plasma Hcy concentrations were significantly increased in a total of 51 patients receiving monotherapy or various combinations of AEDs as compared to controls (4). More recently, Yoo and Hong reported that approximately 25% of patients receiving AEDs had hyperhomocysteinemia (5). Apeland confirmed these observations but demonstrated that AEDs are, by themselves, independent predictors of a high plasma Hcy level (6).

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As to the effect of AEDs therapy on vitamin B₁₂ or vitamin B₆ nutritional status, no consensus has been achieved in the literature (7-10). Furthermore, reports on the effect of AEDs on Hcy concentrations are scarce. Elevated plasma homocysteine concentration has been associated with an increased risk for occlusive vascular disease. Hyperhomocysteinemia may be caused by the deficiency of one of the vitamins or the combination, and these deficiencies have been frequently associated with the administration of AEDs. We hypothesized that hyperhomocysteinemia exists in patients receiving AED; thus, a long-term administration of these drugs possibly results in an increased risk of occlusive vascular disease. To test this hypothesis, we undertook the study presented here to evaluate whether differences exist in Hcy serum concentrations in children receiving VPA, monotherapy, in comparison with healthy control subjects and to determine the possible relationship between Hcy levels and folate, vitamin B₁₂ serum concentrations. For this purpose, we measured serum concentrations of Hcy, folate, and vitamin B₁₂ in outpatients with epilepsy who were treated with VPA and in healthy controls.

MATERIALS AND METHODS

We studied 65 patients, aged from 0,583 to 13 years, suffering from various types of epilepsy (38 tonic-clonic, 15 myoclonic-astatic, 7 tonic, and 5 clonic) and 25 healthy subjects, aged from 0 to 15 years. Duration of the illness (year) was 3.7±1.5 (mean± SEM). They did not suffer from any other disease. Fasting blood samples obtained from

subjects in the morning. After coagulation, the blood samples were centrifuged, separated, and stored at -20 °C on the day of sample collection. Hcy, vitamin B₁₂ and folate were investigated in the serum. Serum tHcy levels were analyzed by enzyme linked immunosorbent assay (ELISA) kit (Homocysteine Enzyme Immunoassay Kit, Bio-Rad Lab, Oslo, Norway). All samples were assayed in duplicate. Vitamin B₁₂ and folate values were measured by Bio DPC kits using Immulite 2000 instrument with Chemiluminescence method. Since the blood samples used in this study were collected for routine blood analysis, additional consent from patients and clearance by the Ethics committee were not required.

Statistical analysis: Data were analyzed by using the SPSS for Windows computing program. Results were expressed as mean ± SEM. P values < 0.05 were regarded as statistically significant.

RESULTS

Brief statistics for homocysteine, folate and vitamin B₁₂ levels are presented in Table 1. Serum Hcy concentrations were significantly higher in epileptic children than in healthy controls (p<0.001). However, serum folate levels were significantly lower in VPA receiving group than in controls (p<0.001). The mean vitamin B₁₂ concentrations were found to be unchanged in patients compared to controls. In correlation analyses, in patients with epilepsy receiving VPA treatment total Hcy had significantly negative correlations with folate levels.

Table 1. Homocysteine, folate and vitamin B₁₂ concentrations in serum from patients with epilepsy and healthy control subjects

	Epilepsy (n=65) mean±SEM	Healthy controls (n= 25) mean±SEM	p
Folate (nmol/L)	5.1 ± 2.4	10.4 ± 3.5	<0.001
Vitamin B ₁₂ (nmol/L)	619 ± 282	799 ± 285	NS
tHcy (µmol/L)	17.6 ± 8.9	9.4 ± 6.5	<0.001
Valproate (µg/mL)	51.2 ± 44.7	---	

DISCUSSION

Data from several investigations demonstrates that VPA is associated with either low or normal folate levels, either high or normal plasma tHcy levels (6, 12-15). A fasting plasma tHcy concentration above 15 mmol/L is the most common definition of hyperhomocysteinemia (16), which is associated with approximately a three-fold increased risk of myocardial infarction. For these reasons, the administration of folate is recommended by Ono (3) in epileptic patients with hyperhomocysteinemia due to folate deficiency to prevent vascular disease. The current study shows that epileptic patients receiving VPA have increased plasma levels of Hcy. Our results are in agreement with a previous report on Hcy concentrations in patients with epilepsy (12).

The increased Hcy concentration may be due to a deficiency in vitamins necessary for the metabolism of Hcy. On the other hand, high plasma levels of Hcy could be related to the presence of epilepsy. However, the baseline evaluation allows excluding the possibility that this abnormality could be a result of the convulsive disorder itself or other situations (e.g. genetic abnormalities of metabolism of Hcy).

In order to better understand the metabolism of Hcy, we also studied the levels of serum vitamin B₁₂ and serum folate. We found significantly lower serum folate concentrations in patients with epilepsy than healthy controls. However, there was no difference in concentrations of vitamin B₁₂ between patients and controls.

The mechanisms by which AEDs alter folate metabolism are still uncertain. Data on VPA effects on folic acid are conflicting. The present study shows that there was a decrease of serum folate levels in patients being treated with VPA. Contrary to our findings, Apeland reports that valproate does not influence folate and Hcy levels in adult patients (6). Wegener and Nau described that VPA

increased the concentrations of folate (17). The effect of valproate on plasma folate concentrations as shown by this study is similar to that by Verrotti (12). Their patients on VPA had high plasma tHcy and low folate levels. The reason for the difference between these findings is unknown. It may be possible to attribute it to the difference in the practice of folic acid fortification for commonly consumed foods between the countries. As all study subjects were from the same cultural and geographic region, we believe that there were only minor dietary differences between patients and controls. However, it may be a correlation between folate levels and drug dosage or duration of treatment. Serum folate levels were not correlated with serum concentrations of VPA. Our data indicate that the result of long-term treatment with VPA may exert a major influence on serum folate levels.

The literature on the vitamin B₁₂ status in patients undergoing treatment with VPA is controversial. Decreased, normal and increased vitamin B₁₂ concentrations have been reported in patients with epilepsy (11-13). We found no difference in concentrations of vitamin B₁₂ between patients and controls. This study did not confirm the previous finding that patients on VPA have elevated vitamin B₁₂ levels in serum (9, 10, 13).

In conclusion, our findings suggest that VPA play a critical role in hyperhomocysteinemia and the folate depletion seen in patients receiving VPA. Thus, a long-term administration of these drug possibly results in an increased risk of occlusive vascular disease.

These results indicate that homocysteine levels should be determined in order to identify hyperhomocysteinemia among patients receiving sodium valproate. These patients may have a higher folate requirement to maintain a normal homocysteine level because of a nutritional deficiency of vitamin B₁₂ or folate leads to increased homocysteine concentrations.

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