Serum Level of Homocysteine, Folate, and Vitamin B12 in Adult Epileptic Patients Under Antiepileptic Therapy

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Abstract

Background:
Epilepsy is a medical condition that requires long-term treatment with antiepileptic drugs. Different epidemiological studies revealed an increased risk of atherogenic cardiovascular diseases in patients under antiepileptic therapy. The possible etiologic suggestion is alterations of the serum homocysteine, folate, and vitamin B_{12} metabolism.

Objective:
To evaluate serum homocysteine, folate, and vitamin B_{12} level in patients under antiepileptic monotherapy.

Methods:
This cross-sectional study was conducted in the Department of Biochemistry, Dhaka Medical College from July 2017 to June 2018 through purposive sampling. In this study, forty diagnosed patients under antiepileptic monotherapy and forty age and sex-matched apparently healthy control were selected according to selection criteria, from OPD of the Neurology department, Dhaka medical college hospital.

Results:
In patients under antiepileptic therapy, mean (±SD) of serum homocysteine (µmol/L), folate (ng/ml), and vitamin B_{12} (pg/ml) were 14.15±3.4, 13.61±2.02, 361.78±41.26 respectively. In the healthy control group, serum homocysteine (µmol/L), folate (ng/ml), and vitamin B_{12} (pg/ml) were 8.78±2.59, 16.71±2.06 and 366.69±44.15 respectively. Serum homocysteine level was found higher and folate level was found lower in patients with antiepileptic therapy which were statistically significant ($p<0.001$). But serum B_{12} level difference was found statistically non-significant. Serum homocysteine level has a positive correlation (p-value 0.001, r value 0.71) and serum folate has a negative correlation (p-value 0.001, r value -0.65) with the duration of antiepileptic therapy.

Conclusion:
A higher level of serum homocysteine and a lower level of folate was found in patients under antiepileptic therapy. But there was no significant change in the serum vitamin B_{12} level of these patients.

Keywords: Homocysteine, Folate, Vitamin B_{12}, Antiepileptics

Introduction:

Epilepsy is a major neurologic disorder that affects about five to ten percent of people worldwide. It is a chronic and dynamic medical condition that requires long-term and usually lifelong treatment with antiepileptic drugs.[1] In Bangladesh, the overall prevalence of epilepsy is 8.6% in urban areas and 7.9% in rural areas.[2] Epidemiological studies have shown that in adults suffering from epilepsy, the risk of development of atherogenic ischemic heart disease (IHD) and fatal cardiovascular disease increase to 34% and 68%, respectively.[3]

The underlying etiology of atherosclerosis-related vascular disease in epileptic patients has not yet been fully addressed. The possible etiologic suggestion is the alteration in the homocysteine metabolisms.[4] Impairment of endothelial function is another possible mechanism, which has been presumed to be the result of epilepsy itself or the side effects of long-term treatment with antiepileptic agents. This may induce the process of atherogenesis and result in arterial obstructive diseases leading to stroke, myocardial infarction, etc.[5] The complex metabolism of homocysteine in the body is highly dependent on vitamin-derived

co-factors and deficiencies in vitamin B_{12} and folic acid are associated with hyperhomocysteinemia.

Homocysteine is associated with damage to the arteries which is thought to interfere with the way cells use oxygen, resulting in a build-up of damaging free radicals. Oxidation causes many diseases, including heart disease, stroke, cancer, and autoimmune diseases. Homocysteine accumulates in the body causing cell damage and the onset of major disease, is because the biochemical transformation process is not working properly, usually due to a lack of these needed vitamins.

Common antiepileptic medications such as carbamazepine, phenytoin, or phenobarbital are recognized as a cause of folate and vitamin B deficiency.[6] In contrast antiepileptic drugs that have various effects on enzyme induction in the liver folic acid, and valproate (VPA) have no effect on hepatic enzyme induction. Further, VPA impairs intestinal absorption of folic acid, and directly interferes with the metabolism of folic acid coenzymes.[7] Therefore it is conceivable that AED treatment increases plasma concentrations of homocysteine.

The study was carried out to assess the effect of monotherapy with common anti-epileptic drugs on homocysteine metabolism. There were also attempts to uncover the relationship between the levels of homocysteine with the cofactors involved in its metabolism.

Methods:

This cross-sectional analytic study was carried out in the department of Biochemistry, Dhaka Medical College during the period from July 2017 to June 2018 through purposive sampling. Forty adult epileptic patients under antiepileptics monotherapy and age and sex-matched another forty healthy were selected according to selection criteria from the Neurology outpatient department of Dhaka Medical College Hospital. Inclusion criteria were diagnosed cases of epilepsy confirmed by a Neurologist, and patients under antiepileptic drugs monotherapy therapy within the therapeutic dose range for at least six months. Exclusion criteria were vitamin B12 or folic acid therapy, previous history of stroke and ischaemic heart disease, chronic kidney disease, chronic liver disease, chronic obstructive pulmonary disease, malignancy, pregnancy and lactation, and drugs other than antiepileptic that increase serum homocysteine level (Levo-dopa, Methotrexate, Theophylline, Penicillamine, Isoniazid, Hydralazine, and Oral Contraceptive Pill). Informed written consent was taken from the study subjects and ethical approval was obtained from the ethical committee of Dhaka Medical College. Age, sex, BMI, and BP were recorded. With all aseptic precautions, 5 ml of venous blood sample was collected from each study subject. Serum homocysteine, folate, and vitamin B12 were estimated by an automated analyzer: Architect plus ci 4100. All data were recorded in a pre-designed data collection form. Continuous variables were expressed as mean ± SD and were compared between groups of patients by unpaired Student's 't' test. Qualitative data were analyzed by Chi-square test. The level of significance was defined as p-value <0.05 at a 95% confidence interval. To observe correlation, Pearson's correlation coefficient test was done. All analysis was done using the SPSS version 22.

Result:

The Mean (±SD) age of patients with antiepileptics (Group A) was 26.80±5.92 years and the control group (Group B) was 26.98±5.33 years (p=0.89) which showed a statistically non-significant difference (Table-I).

Table-I: Distribution of age and sex in the study subjects (n=80)

Demographic features	Group A (n=40)	Group B (n=40)	p-value
Age (years) Mean ± SD	26.80±5.92	26.98±5.33	0.89
Gender			
Male	22	19	0.66
Female	18	21	

Unpaired students' "t" test and chi-square test were done to measure the level of significance; significance= (p<0.05).

Unpaired students' "t" test and chi-square test were done to measure the level of significance; significance= (p<0.05).

There was statistically no significant difference found among the study groups with respect to BMI, systolic, and diastolic blood pressure (p>0.05) (Table II).

Table-II: Clinical parameters of the study subjects (n=80)

Clinical parameters	Group A (n=40) Mean±SD	Group (n=40) Mean±SD	p-value
BMI (kg/m^2)	24.85±1.34	24.81±1.24	0.91
Systolic BP (mm of Hg)	116±6.32	120.25±9.2	0.09
Diastolic BP (mm of Hg)	74.25±5.26	76.13±5.72	0.58

Unpaired student's 't-test was done to measure the level of significance; significance= (p-<0.05)

Mean ±SD of serum homocysteine level increased and folate level decreased in patients under antiepileptic therapy than the control group which was statistically significant (p<0.05) but both were within normal limits. The mean ±SD of serum B_{12} level also decreased in patients under antiepileptic therapy than the control group, which was statistically not significant (p>0.05) (Table III).

Table-III: Serum homocysteine, folate and vitamin B_{12} level in the study groups (n=80)

Parameters	Group A (n=40) mean±SD	Group B (n=40) mean±SD	p-value
Homocysteine (µmol/L)	14.15±3.4	8.78±2.59	0.001
Folate (ng/ml)	13.61±2.02	16.71±2.06	0.001
Vitamin B_{12} (pg/ml)	361.78±41.26	366.69±44.15	0.71

(Normal serum value: Homocystine: 5-15µmol/L 8 , Folate: 2–20 ng/ml 9, Vitamin B 12:200-900 pg/ml 9)

Unpaired student's test was done to measure the level of significance; significance= (p<0.05)
There was a significant positive correlation between serum homocysteine level and the duration of antiepileptic therapy (Figure-1).

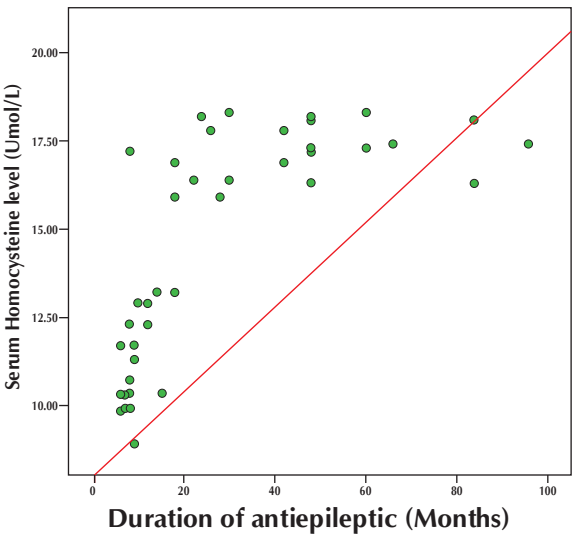

Figure-1: Correlation between serum homocysteine level and duration of antiepileptic therapy in months

There was a significant negative correlation between serum folate level and the duration of antiepileptic therapy (Figure-2).

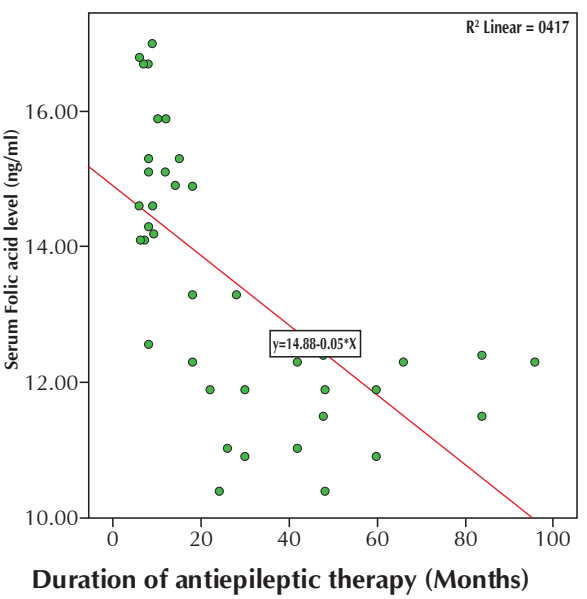

Figure-2: Correlation between serum homocysteine level and duration of antiepileptic therapy in months

Discussion:

In our study, we have found mean (±SD) serum homocysteine levels were 14.15 ± 3.4 and 8.78 ± 2.59 µmol/L in A and B groups respectively. So the

mean value of serum homocysteine level was found to increase in patients under antiepileptic therapy than in the control group, which is statistically significant ($p<0.05$). Schwaninger et al[8] did another cross-sectional study and found serum homocysteine level 14.7 ± 3 and 9.5 ± 0.5 µmol/L in both groups respectively and was significantly increased in the A group. Sener et al[9] also found significantly increased serum homocysteine levels in the A group. In the study, serum homocysteine level was 17.9 ± 1.41 and 9.8 ± 0.59 µmol/L in A and B groups respectively. Other studies done by Paknahad et al[10], Dayem et al[11], Eldeen et al[12], Kurul et al[13] also found significantly increased serum homocysteine levels in patients with antiepileptic drug therapy. So all of these studies are supportive of our current study findings.

According to our study, we have found mean (±SD) serum folate levels were 13.61 ± 2.02 and 16.71 ± 2.06 ng/ml in A and B groups respectively. So the mean value of serum folate level was found to decrease in patients under antiepileptic therapy than in the B group, which is statistically significant ($p<0.05$). Schwaninger et al[8] found serum folate levels 13.5 ± 1 and 17.4 ± 0.8 ng/ml in the cross-sectional study respectively and were significantly decreased in the case group. Sener et al[9] also found significantly decreased serum folate levels in the case group. Other studies done by Paknahad et al[10], Dayem et al[11], Eldeen et al,[12] Kurul et al[13] also found significantly reduced serum folate levels in patients with antiepileptic drug therapy. All of these studies' findings are consistent with our current study findings.

In our current study, we have found mean (±SD) serum vitamin B_{12} levels were 361.78 ± 41.26 and 366.69 ± 44.15 pg/ml in A and B groups respectively. So the mean value of serum vitamin B12 level was found to decrease in patients under antiepileptic therapy than in the control group, which is statistically non-significant ($p=0.71$). Schwaninger et al[8] in a cross-sectional study found serum vitamin B12 levels 363.7 ± 39.8 and 368.5 ± 36.1 pg/ml in both groups respectively and the difference was not significant. Other studies done by Paknahad et al[10], Dayem et al[11], Eldeen et al,[12] Kurul et al[13] found no significant difference among both groups with respect to vitamin B12 level. These studies' findings are also consistent with our current study findings.

Our study shows a positive correlation between serum homocysteine level and the duration of antiepileptic therapy. Pearson's correlation coefficient test revealed a positive correlation coefficient ($r= 0.71$) which is statistically significant ($p=0.001$). Therefore, it can be concluded that serum homocysteine levels will increase with an increased duration of antiepileptic therapy. This result is consistent with Eldeen et al[12], who also found a positive correlation between serum homocysteine level and duration of antiepileptic therapy.

According to the study negative correlation was observed between serum folate level and the duration of antiepileptic therapy. Pearson's correlation coefficient test revealed a negative correlation coefficient ($r = -0.65$) which is statistically significant ($p=0.001$). Therefore, it can be concluded that serum folate levels will decrease with an increased duration of antiepileptic therapy. Eldeen et al[12] also found a negative correlation between serum folate level and duration of antiepileptic therapy.

A higher level of serum homocysteine and a lower level of folate were found in patients under antiepileptic therapy. But there was no significant change in the serum vitamin B_{12} level of these patients. So routine periodic estimation of serum homocysteine and folate level may be advocated for patients with antiepileptic therapy to prevent hyper-homocysteinemia-related complications.

Conclusion:

A higher level of serum homocysteine and a lower level of folate were found in patients under antiepileptic therapy. But there was no significant change in serum vitamin B 12 level of these patients. So routine periodic estimation of serum homocysteine and folate level may be advocated for the patients with antiepileptic therapy to prevent hyper-homocysteinemia related complications such as ischemic heart disease, stroke, neural tube defect of fetus in pregnant women.

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