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Short Communication

Serum folate and homocysteine levels in patients with obsessive-compulsive disorder

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Abstract Previous studies have shown that folate deficiency, increased homocysteine, impaired metylation have been identified in depressive disorder. Recently, growing research has resulted in the biological association between obsessive-compulsive disorder (OCD) and affective disorders. Therefore, in the present study it was evaluated whether or not folate and homocysteine levels changed. Serum folate and homocysteine concentrations were measured in 23 patients with OCD and in same number of controls. In addition, all patients were assessed by Yale-Brown Obsession Compulsion Scale (Y-BOCS). Serum folate values were significantly lower in OCD patients than in controls, while homocysteine concentrations were higher in patients compared with controls. Serum folate values were significantly and negatively related to Y-BOCS scores. Total serum homocysteine concentrations were positively correlated to Y-BOCS scores and the duration of illness. There was a trend toward a negative correlation between the concentrations of serum folate and homocysteine. In conclusion, we identified that a group of patients with OCD might have folate deficiency, higher homocysteine levels and probable impaired metylation and monoamine metabolism.

Key words folate, homocysteine, metylation, obsessive-compulsive disorder.

INTRODUCTION

Earlier reports suggested that there was a relationship between folate deficiency and psychiatric illnesses, especially depressive disorders.[1] Recent trials have found that total plasma homocysteine seems to be a sensitive marker of functional deficiency of folate.[2,3] The synthesis of methionine from homocysteine requires a supply of methyl groups from methyl folate. Methyl folate is needed for the production of S-adenosyl methionine (SAM). SAM serves as the main methyl donor in the brain for the synthesis of amines, neurotransmitters, nucleoproteins and membrane reactions. It was reported that both SAM and methyl folate appeared to have an antidepressant effect and a potential to switch from depression to mania.[4] In addition, Carney reported that folate was correlated with cerebrospinal fluid (CSF) 5-hydroxy indole asetic acid (5-HIAA).[5]

The contemporary knowledge indicates that obsessive-compulsive disorder (OCD) has a multifactorial etiopathogenesis. Recently, there has been an important advance in the neurobiology of OCD. It has been established that severe brain traumas, impairments in biochemical neurotransmission and receptor functions may be associated with the occurrence of OCD. The comorbidity of OCD and depressive disorder is an observation that is frequently seen. OCD has frequently depressive symptoms and comorbid depressive disorder, on the other hand, obsessions and compulsions may be observed in the patients with depressive disorder.[6,7] Recently, this relationship resulted in a large body of research regarding the biological association between OCD and affective disorders. In our previous study we suggested that OCD might be a heterogeneous subtype including some biological indica-

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tions of anxiety and affective disorders.[8] To the best of our knowledge, there has been only one study in the literature regarding folate but not homocysteine in OCD. Therefore, we carried out a study to specifically measure serum folate and homocysteine levels to see whether or not its eventual alterations might have an etiopathogenetic significance in patients with OCD.

METHODS

The sample consisted of 23 patients who attended to Firat University Scool of Medicine Department of Psychiatry and who were diagnosed with pure OCD according to DSM-IV and who met the admission criteria. A DSM-IV diagnosis of OCD was established on the basis of independent clinical interviews by one senior psychiatrist. All subjects gave informed written consent and the study had been approved by local ethic committee in accordance with the Declaration of Helsinki.

Twenty-three healthy control subjects (mean age = 28.4 ± 7.5 years; range 21–45) were chosen from the hospital staff according to exclusion criteria. Controls were interviewed with the non-patient version of the SCID (SCID-NP) to exclude any axis I disorder.[9] In addition, controls did not a have history of major mood disorder, dementia, mental retardation or psychosis in their first-degree relatives. Controls were matched with the patients in regard to sex and age.

All subjects had either no history of treatment or were drug-free for at least the last 2 weeks. They underwent physical examination, total biochemistrical evaluation, chest X-ray, urinalysis and ECG. Liver and kidney function tests were evaluated. All participants were carefully assessed to rule out autoimmune, pulmonary, infectious diseases and neoplasms. Exclusion criteria were the history of any endocrinological condition, women who are pregnant or lactating, or of childbearing potential, clinically relevant abnormal laboratory tests including megaloblastic anemia, alcohol abuse and dependence, and the history of immunologic disease. The patients were administered Yale-Brown Obsession Compulsion Scale (Y-BOCS)[10] to determine the severity of OCD. Data on smoking were obtained from each patient using a questionnaire, 1 day before blood drawing. Smoking was not permitted after 23:00 hours, 1 day before blood drawing.

Blood sampling and determination of folate and homocysteine

Venous blood samples from the left forearm vein were collected into 5 mL vacutainer tubes containing potassium EDTA between 07:00 and 08:00 hours after overnight fasting. Some hematological parameters were measured using an autoanalyzer (Coulter Max M, Coulter Electronics Ltd, Luton, UK). The blood samples were centrifuged at 4000 r.p.m. for 10 min at 4°C to remove plasma. Folate values were measured by using folic acid (Roche Diagnostics Elecsys module E170 hormone autoanalyzer, Mannheim, Germany).[11] Homocysteine levels were measured by Axis Homocysteine kit (ELISA) (Axis-Shield Group Rodeløkka, Oslo, Norway).[12]

Statistical analysis

Obtained data were evaluated by SPSS for windows 9.0 (1998; SPSS Inc, Chicago, IL, USA). The comparisons within group were performed using Wilcoxon's rank sum test, while those between intergroups were carried out by using independent samples *t*-test. The χ^2 test was used to compare the proportion of non-suppression between groups. For correlation evaluations, the Spearman correlation (two-tailed) test was used. $P < 0.05$ was considered to be significant.

RESULTS

A total of 23 patients (15 females and eight males) with a mean age of 29.1 ± 6.3 years (range 18–44) were enrolled in this study. The control group ($n = 23$) had 13 females and 10 males, with a mean age of 27.2 ± 5.4 years, ranged 21–44 ($P > 0.05$). There were no significant differences in age or in female/male ratio between the patients and controls ($P > 0.05$). Thirteen patients and 10 the controls were smokers. Eleven of 13 smoker patients had >20 cigarettes per day except for two patients who had between 10 and 20 cigarettes per day. In contrast, all 10 smoker controls had >20 cigarettes per day. The mean duration for smoking in patients and controls was 8.2 ± 4.8 and 6.9 ± 4.1 years, respectively ($P > 0.05$).

There were no statistically significant differences between hematological parameters of the patient groups and controls ($P > 0.05$).

Five patients (21.7%) with mild OCD, nine (39.1%) with moderate OCD, seven (25.9%) with severe and two patients (13.3%) with significant severe OCD were determined by Y-BOCS. The patients had a mean Y-BOCS score of 23.1 ± 5.9. Prior to being entered to the present study, they received various classes of psychotropic drugs: selective serotonin reuptake inhibitor ($n = 17$), clomipramine ($n = 15$), typical neuroleptics ($n = 12$), lithium ($n = 11$), clonazepam ($n = 6$) and buspirone ($n = 3$).

The mean serum folate level was within the normal range in both the patient and controls, with close to

below range in the patient group. The level was lower in patients compared to control subjects (3.4 ± 2.8 *vs.* 7.9 ± 3.9 ng/mL; $P < 0.01$). Seven patients (30.4%) from the patient group and no one from controls had low serum folate levels when taking into consideration normal folate levels. Only one patient and two controls had high serum folate according to the upper values of normal folate levels. Serum folate levels were minimally higher in smokers than in non-smokers but the difference was not statistically significant in patients with OCD (smoker, 3.5 ± 3.1 ng/mL; non-smoker, 3.2 ± 2.4 ng/mL; $P > 0.05$) and in the controls (smoker, 8.0 ± 4.4 nmol/L; non-smoker, 7.8 ± 3.2 nmol/L; $P > 0.05$). Folate levels were also unaffected by gender in both patient and control groups ($P > 0.05$). The serum folate and homocysteine levels in patients and controls are shown in Fig. 1.

In both patient and control groups, homocysteine levels were within the normal range. However, the level was higher in patients compared to control subjects (21.8 ± 8.9 *vs.* 8.3 ± 5.1 μmol/L; $P < 0.01$). Eight OCD patients (34.8%) and two controls (8.7%) had a total serum homocysteine concentration above the normal range. The effects of smoking and gender were examined in both patients and controls. Homocysteine levels were unaffected by smoking in both patient (smoker, 20.9 ± 7.8 ng/mL; non-smoker, 22.9 ± 9.4 ng/mL; $P > 0.05$) and control groups (smoker, 8.5 ± 4.5 ng/mL; non-smoker, 8.1 ± 5.5 ng/mL; $P > 0.05$). There were also no effects of gender on the homocysteine in this study in both patient and control groups ($P > 0.05$).

Serum folate values were significantly and negatively related to Y-BOCS scores ($r = -0.51$, $P < 0.05$) but not related to age, previous psychotropic medication and the duration of illness. Total serum homocysteine concentrations were positively correlated to Y-BOCS scores ($r = 0.55$, $P < 0.05$) and the duration of illness ($r = 0.55$, $P < 0.05$) but not related to age, and previous psychotropic medication. There was a trend toward a negative correlation between the concentrations of serum folate and homocysteine ($r = -0.50$, $P < 0.05$).

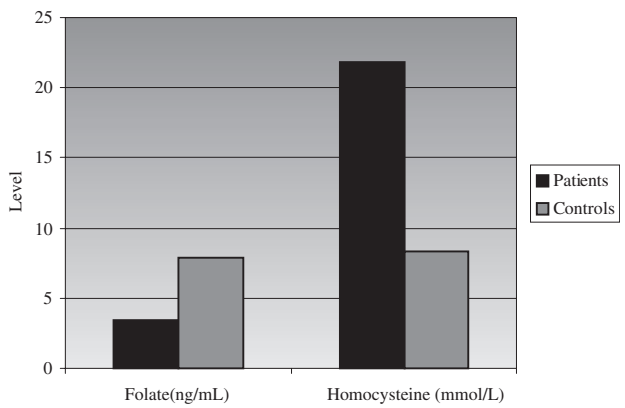

Figure 1. Serum folate and homocysteine levels in patients and controls.

DISCUSSION

To the best of our knowledge, this is the second study regarding folate and the first one regarding homocysteine values in patients with OCD. The data we found provide evidence that serum folate levels decrease and homocysteine levels increase in patients with OCD. Apart from this, serum folate values were significantly and negatively related to Y-BOCS scores, while there was a positive correlation between the total serum homocysteine concentrations and Y-BOCS scores or the duration of illness. These findings do not agree with Hermesh *et al.*, who indicated that serum vitamin B_{12} levels were associated with some OCD but folate levels did not.[13]

It has been proposed evidence concerning the role of folate in the central nervous system function because of the essential role of folate in the one-carbon cycle that furnishes SAM, the principal methyl donor for a broad range of reactions involving the synthesis of neuroactive substances, the formation of membrane phospholipids, and the metabolism of nucleic acids.[14] When administered in parenteral and certain oral forms, both SAM and methyl folate have been shown to have antidepressant efficacy greater than placebo and comparable to that of tricyclic antidepressants[15] and are involved in the switching from depression to mania.[4] Folate also appears to influence the rate of synthesis of tetrahydrobiopterin,[16] a cofactor in the hydroxylation of phenylalanine and tryptophan, rate-limiting steps in the biosynthesis of dopamine, norepinephrine, and serotonin, neurotransmitters postulated to play a role in the monoamine hypothesis of affective disorders. In addition, methyl tetrahydrofolate has been shown to bind to presynaptic glutamate receptors,[17] where it may potentially modulate the release of other neurotransmitters, including the monoamines. Folate is important in the synthesis of tetrahydrobiopterin, which is the cofactor for the hydroxylation of phenylalanine and tryptophan and is the rate-limiting step in the synthesis of dopamine, norepinephrine and serotonin. According to the monoamine hypothesis of affective disorders, depression is due to a deficiency of 5-HT, or norepinephrine, or both these monoamines. Investigation of depressive patients showed an abnormality of tetrahydrobiopterin in these patients and that this was related to plasma folate.[18] Low cerebrospinal fluid levels of the serotonin metabolite 5-hydroxyindole acetic acid and the dopamine metabolite homovanillic acid (HVA)

have been reported in several, although not all, studies among folate-deficient patients with epilepsy, other neuropsychiatric disorders, and congenital folate-deficiency states.[19–22] The studies dealing with the changes in the monoamine and its metabolites levels in biological fluids of patients with OCD demonstrated inconsistent findings. Some studies reported high levels of epinephrine, 5HT and HVA,[23] others reported low levels.[24] It may be that homocysteine is a very good index of folic acid status of the patient or that it has, a yet unspecified interaction with the CNS that produces depression. It has now been established in many investigations that folic acid will lower plasma homocysteine and in contrast, decreased folate levels will elevate homocysteine levels, and so elevated levels of homocysteine, resulting from folate deficiency, may play a role in mediating some of its neuropsychiatric complications by both generating elevated levels of S-adenosyl-homocysteine, which broadly inhibits methylation reactions, and also possibly exerting direct excitotoxic effects via activity at the N-methyl-D-aspartate glutamate receptors.[25,26] Meanwhile, we should note that positive correlation between the plasma concentration and cerebrospinal fluid concentrations for both folate and homocysteine was determined.[27] OCD has frequently depressive symptoms and comorbid depressive disorder, on the other hand, obsessions and compulsions may be present in patients with depressive disorder.[6,7] Moreover, according to most recent hypotheses, OCD may be the consequence of a developmental pathology affecting serotonergic and/or dopaminergic neuronal systems which also have been suggested for the etiopathogenesis of depression.[28] Therefore, these relationships have resulted in growing research about the biological association of OCD and affective disorders, as supported by the present paper.

In conclusion, the present study suggests that OCD may be associated with folate and homocysteine but our sample is too small to allow us to conclude that this alteration may be an important biological indicator for this disorder.

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