THE RELATIONSHIP BETWEEN GLAUCOMA AND SERUM VITAMIN B12, FOLIC ACID LEVELS AND NUTRITION

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Abstract

Aims: To examine the relationship between diet, serum vitamin B12 and folic acid levels and glaucomatous optic neuropathy.

Materials and methods: The study included 77 glaucoma patients and 40 healthy individuals as the control group. Vitamin B12 and folic acid levels in serum were measured by chemiluminescence method. Nutrition and diet experts analyzed dietary intake vitamin B12 and folic acid levels of all individuals in the study by using a questionnaire.

Results: Median and standard deviation of vitamin B12 serum levels were 231 and 135.19 in glaucoma patients. They were 272.5 and 84.54 in the control group, and the difference was statistical significant (p=0.016). Median and standard deviation of serum folic acid levels were 7.86 and 3.15 in glaucoma patients and were 6.9 and 3.1 in the control group. The difference was not statistical significant (p=0.144). Analysis of vitamin B12 and folic acid levels of glaucoma patients with serum vitamin B12 levels according to their food consumption revealed that 1 of 16 glaucoma patients had normal vitamin B12 intake, while 15 had deficient vitamin B12 intake. Dietary intake of folic acid was found to be deficient in 14 of 16 glaucoma patients.

Conclusion: Determination of low serum level of vitamin B12 in 16 of 77 glaucoma patients may indicate the importance of dietary vitamin B12 intake in glaucoma patients.

Key words: Glaucoma, vitamin B12, folic acid, nutrition.

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Introduction

Glaucoma is a chronic, progressive optic neuropathy with ganglion cell damage and visual field loss that can result in irreversible blindness. Degeneration of retinal ganglion cells causes cavitation and atrophy in optic nerve head⁵. Glaucoma affects more than 67 million people worldwide. Approximately 10% of these people are estimated to be blind. It ranks as a third cause of blindness and account for approximately 14% of all blindness cases. While approximately 50% of glaucoma cases are identified in developed countries, this ratio is lower in developing ones⁶⁷.

The exact mechanism causing ganglion cell damage in glaucoma is still unclear. Age, race, family history, diabetes, high intraocular pressure (IOP), myopia, pseudoexfoliation increased “cup/disc” (C/D) ratio in optic nerve head and low diastolic perfusion pressure are the known as important risk factors⁸⁻¹¹.

The most common types of glaucoma are primary open angle glaucoma (POAG), pseudoexfoliative glaucoma (PEXG) and normotensive glaucoma (NTG). IOP is considered as the most important risk factor in glaucoma.

Recently, it was shown that vascular risk factors can also have a role.
Distorted microcirculation and abnormal perfusion can cause glaucomatous damage in optic nerve head\(^{(12)}\).

Vitamin B12 and folic acid (vitamin B9) have a prominent role in development, differentiation and function of central nervous system. These vitamins play a role in methionine-homocysteine pathway, which is responsible for the supply of methyl groups that are needed for DNA and protein synthesis. Methionine becomes activated and turns into S-adenosylmethionine (SAM), which is responsible for methylation of numerous important molecules. S-adenohomocysteine (SAH) is formed by the separation of methyl group from SAM. The increase in SAH concentration disturbs a series of metabolic processes in brain by decreasing SAM-dependent methylation capacity through antagonistic mechanisms. Cellular function losses, DNA damage and impaired myelin synthesis can be shown as an example to these processes. SAH turns into homocysteine after hydrolysis\(^{(13)}\).

Homocysteine is a neuro-vascular-toxic by-product that contains sulfur. Homocysteine can turn into firstly cystatione and then to cystine by transulfuration. Cysteine is a glutathione component. Deficiency of vitamin B12 or folic acid decreases re-methylation of homocysteine to methionine, which results in elevation of homocysteine level. Decreased SAM synthesis causes hypomethylation. This impairs the synthesis of various important molecules including protein and neurotransmitters that are important for structural integrity of the brain\(^{(13)}\). Although in recent years it was reported that plasma homocysteine level increases in POAG and PEXG, it was also reported no significant difference in plasma homocysteine levels in POAG and NTG\(^{(12)}\).

Vitamin B12 is found in animal products such as meat, milk and cheese. It is an essential vitamin for the continuation of the vitality of blood and optic cells. Optic neuropathy especially with prominent central nervous system and hematologic findings can occur in deficiency of vitamin B12. Pale optic disc, optic disc change similar to glaucomatous optic atrophy, bilateral symmetric central visual field loss and discromatopsia are optic neuropathy findings associated with vitamin B12 deficiency. Homocysteine increase due to vitamin B12 and folic acid deficiency is held responsible for neurodegenerative diseases such as Alzheimer’s and Parkinson’s disease\(^{(16)}\).

The aim of this study is to investigate the diet of glaucoma patients and the control group; comparison of serum vitamin B12 and folic acid levels and to examine the relationship between diet, serum vitamin B12 and folic acid levels and glaucomatous optic neuropathy.

Materials and methods

The study included 77 glaucoma patients having C/D ratio over 0.5 and significant glaucomatous damage in visual field and 40 healthy individuals as the control group. The patients were classified according to visual fields examples as moderate and severe\(^{(15)}\). The individuals who underwent gastrointestinal system surgery or have gastrointestinal system disease affecting vitamin B12 and folic acid deficiency; receiving vitamin B12 and folic acid treatment; having ocular disease apart from glaucoma and hematologic diseases were excluded from the study. Eye examination of all participants was performed. Anterior chamber and posterior segment were examined by the Goldman three mirror lenses. IOPs were measured by the Goldman applanation tonometer. Central corneal thicknesses were measured by the ultrasonic pachymeter. Visual field tests were performed by the Humphrey Field Analyzer, SITA standard, 24-2. The best corrected visual acuities, C/D ratios and IOPs were evaluated.

Vitamin B12 and folic acid level in serum was measured by chemiluminescence method (IMMULITE 2500, Diagnostic Products Corporation, USA). According to used chemiluminescence method, the patients having vitamin B12 below 197 pg/ml in serum and those having folic acid below 3 ng/ml were considered as deficient. Those having low vitamin levels in serum were also evaluated by an internal medicine expertise.

Nutrition and diet experts, using a questionnaire, analyzed Vitamin B12 and folic acid levels in food consumption of all individuals in the study. Food consumption of individuals in three successive days were determined and evaluated by “Diet Information System (Bebis)"\(^{(16, 17)}\). The informed consent was obtained from all participants.

All data were analyzed by the Statistical Package for the Social Sciences software (SPSS) 15.0 (Ondokuz Mayis University, trial version). All data were statistically analyzed by the Mann-Whitney U test, Chi-square and Fisher’s Exact test were used for evaluation of categorical variables.
Results

The study included a total of 77 glaucoma patients (41 male, 36 of female). Mean age was 63.2±10.8, C/D ratio was 0.7±1.7 and mean IOP was 14.7±7.4. Control group consisted of 40 people (15 male, 25 female). Mean age was 54.2±11.4, C/D ratio was 0.2±2.8 and mean IOP was 13.9±2.7 (Table 1). There was statistical significantly difference between mean ages of two groups (p<0.001). However, there was no significant difference between C/D ratios of two groups (p=0.2237) and between IOP of two groups (p=0.4902). MD was -5.04±9.81 in the patients and was -0.02±1.7 in the control group. There was a significantly difference (p=0.001) (Table 1). SD was 4.22±4.39 in the patients and was 2.74±2.1 in the control group, with a statistical significant difference (p=0.037) (Table 1).

In the group 1 (patients group), the lowest vitamin B12 level was 150 pg/ml, the highest was 906 pg/ml. Median and standard deviation of vitamin B12 serum levels were 231 and 135.19 in glaucoma patients. In the group 2 (control), the lowest vitamin B12 level was 231 and 135.19 in glaucoma patients. Of the 77 glaucoma patients who were included in the study, 15 were NTG, 36 were POAG and 26 were PEXG patients. It was found that 16 patients had low vitamin B12 serum levels. Of these 16 patients, 2 were NTG, 9 were POAG and 5 were PEXG. It was found that animal protein intake of these 16 patients were insufficient.

Table 1: Demographic data, cup/disc ratio and intraocular pressure of the study groups.

<table>
<thead>
<tr>
<th></th>
<th>Group 1 (Patients with glaucoma)</th>
<th>Group 2 (Control)</th>
<th>p value</th>
</tr>
</thead>
<tbody>
<tr>
<td>No of cases</td>
<td>77</td>
<td>40</td>
<td></td>
</tr>
<tr>
<td>Male/female</td>
<td>41/36</td>
<td>15/25</td>
<td></td>
</tr>
<tr>
<td>The mean age</td>
<td>63.2 ± 10.8</td>
<td>54.2 ± 11.4</td>
<td>&lt;0.001*</td>
</tr>
<tr>
<td>C/D ratio</td>
<td>0.7 ± 1.7</td>
<td>0.2 ± 2.8</td>
<td>0.2237</td>
</tr>
<tr>
<td>The average IOP</td>
<td>14.7 ± 7.4</td>
<td>13.90 ± 2.7</td>
<td>0.4902</td>
</tr>
<tr>
<td>MD</td>
<td>-5.04 ± 9.81</td>
<td>-0.02 ± 1.7</td>
<td>0.001*</td>
</tr>
<tr>
<td>SD</td>
<td>4.22 ± 4.39</td>
<td>2.74 ± 2.1</td>
<td>0.037*</td>
</tr>
</tbody>
</table>

In the group 1 (patients group), the lowest folic acid level was 1.92 ng/ml, the highest was 15 ng/ml. Median and standard deviation of folic acid serum levels were 7.86 and 3.15 in glaucoma patients. In the group 2 (control), the lowest folic acid level was 1.91 ng/ml, the highest was 15 ng/ml. Median and standard deviation of folic acid serum levels were 6.9 and 3.1 in the control group. There was no statistical significant difference (p=0.144) (Table 2).

Table 2: Comparison of serum vitamin B12 and folic acid levels between glaucoma patients and control group.

<table>
<thead>
<tr>
<th></th>
<th>Vitamin B12</th>
<th>Folic acid</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Median</td>
<td>Standard deviation</td>
</tr>
<tr>
<td>Patients</td>
<td>231</td>
<td>135.19</td>
</tr>
<tr>
<td>Control</td>
<td>272.5</td>
<td>84.54</td>
</tr>
</tbody>
</table>

Table 3: Distribution of serum levels of vitamin B12 and folic acid in glaucoma patients and control group.

<table>
<thead>
<tr>
<th></th>
<th>Vitamin B12</th>
<th>Pathological&lt;197 pg/ml</th>
<th>Folic acid</th>
<th>Normal &gt;3 ng/ml</th>
<th>Pathological&lt;3 ng/ml</th>
</tr>
</thead>
<tbody>
<tr>
<td>Glaucoma patients (n=77)</td>
<td>61 (%79.2)</td>
<td>16 (%20.7)</td>
<td>76 (%98.7)</td>
<td>76 (%98.7)</td>
<td>1 (%1.2)</td>
</tr>
<tr>
<td>Control group (n=40)</td>
<td>38 (%95)</td>
<td>2 (%5)</td>
<td>39 (%97.5)</td>
<td>39 (%97.5)</td>
<td>1 (%2.5)</td>
</tr>
</tbody>
</table>

Vitamin B12 serum level was normal in 79.2% (61 patients) of glaucoma patients and low level in 20.7% (16 patients). In the control group, serum vitamin B12 levels were normal in 95% (38 participants) and low in 5% (2 participants). Serum folic acid level was normal in 98.7% (76 patients) and low in 1.2% (1 patient) of glaucoma patients. In the control group, serum folic acid levels were normal in 97.5% (39 participants) and low in 2.5% (1 participant) of the participants (Table 3).
Dietary folic acid intake was found to be deficient in 14 of 16 glaucoma patients (Table 4).

Of the 77 glaucoma patients, 48 were moderate glaucoma patients and 29 were severe glaucoma patients. Of the 16 vitamin B12 deficient patients, 11 were moderate glaucoma patients and remaining 5 were severe glaucoma patients. In addition, of the 61 glaucoma patients with normal vitamin B12 levels, 43 were moderate glaucoma patients and 18 were severe glaucoma patients.

Average of central corneal thickness was 544±17 μm in patients group, while it was 546±13 μm in control group (p>0.05).

Comparison of visual acuities and C/D ratios of glaucoma patients with vitamin B12 deficiency showed that, of the 16 glaucoma patients, best correct visual acuity (BCVA) was >20/40 in 11 and 20/200 and below in 5. Of the control group with vitamin B12 deficiency, best correct visual acuity was 20/20 in 2 individuals. C/D ratio was >0.7 in 11 of glaucoma patients and 0.5-0.6 in 5 glaucoma patients. C/D ratio was 0.5 in one individual in control group (Table 5).

<table>
<thead>
<tr>
<th>Visual acuity</th>
<th>Glaucoma (n=16)</th>
<th>Control (n=2)</th>
<th>C/D ratio</th>
<th>Glaucoma</th>
<th>Control</th>
</tr>
</thead>
<tbody>
<tr>
<td>&lt; 20/200</td>
<td>5</td>
<td>-</td>
<td>0.1-0.4</td>
<td>-</td>
<td>1</td>
</tr>
<tr>
<td>40/200-80/200</td>
<td>-</td>
<td>-</td>
<td>0.5-0.6</td>
<td>5</td>
<td>1</td>
</tr>
<tr>
<td>&gt; 20/40</td>
<td>11</td>
<td>2</td>
<td>&gt; 0.7</td>
<td>11</td>
<td>-</td>
</tr>
</tbody>
</table>

Table 5: Comparison of visual acuities and C/D ration of glaucoma patients with vitamin B12 deficiency and the control group.  
C/D: cup/disc ratio

Of the 16 patients having vitamin B12 level <197 pg/ml in glaucoma patient group, at least one eye of the 5 patients had functional visual loss and bilateral glaucomatous optic neuropathy. In 2 people in control group with vitamin B12 level <197 pg/ml, C/D ratio was 0.5 in 1 individual, visual acuity and visual field were normal.

Other vitamin intake levels of the 16 patients with low vitamin B12 serum level and dietary intake were also evaluated. In this patient group, vitamin B12 intake with diet was normal in 1 patient and low in the other 15. It was found that vitamin B2 intake was low in 9 patients; vitamin B3 intake was low in 6 patients; vitamin B6 level was low in 15 patients; vitamin B9 intake was low in 14 patients; biotin intake was low in 8 patients; pantothenic acid intake was low in 14 patients, vitamin A intake was low in 6 patients; vitamin D and vitamin E intake were low in 16 patients.

Discussion

Unlike other optic neuropathies, glaucoma does not only cause ganglion cell damage but also damages glial tissue. In glaucoma, which is classically defined as optic neuropathy sensitive to IOP increase, elevated IOP is believed to be the most important cause of optic nerve damage. However, some of previous studies observed that IOP level was never over normal value in approximately 20% of the patients with glaucomatous optic nerve damage. Therefore, other mechanisms are believed to be present in pathophysiology of glaucomatous optic neuropathy part from IOP increase. These mechanisms are mechanical theory, ischemic theory and apoptosis theory (2). On the other hand, dietary insufficiency resulting in metabolic optic neuropathy includes important nutrients and especially vitamins such as vitamin B12, vitamin B1, vitamin B2 and folic acid (18).

The literature contains studies on the relationship between glaucoma and homocysteine, vitamin B12 and folic acid. Cumurcu et al. (12) compared serum homocysteine, vitamin B12 and folic acid levels in POAG, PEXG and NTG patients and healthy control group. They reported that homocysteine concentration was significantly higher in patient with PEXG than other groups; however there was no significant difference between other groups. It was reported that vitamin B12 levels did not show a significant difference in all groups; however, folic acid level was significantly lower in patients with PEXG.

Roedl et al. (19) evaluated vitamin B12, vitamin B6 and folic acid levels associated with homocysteine to analyze the cause of hyperhomocysteinemia in patients with PEXG. Comparison of vitamin B12, vitamin B6 and folic acid levels with the control group revealed that the values were significant.
ly lower in patients with PEXG. They reported that this can help to explain the cause of hyperhomocysteineemia in this patient group. Tranchina et al.\(^\text{[20]}\) compared plasma homocysteine, vitamin B12 and folic acid levels of POAG, PEXG and healthy control group. They found that homocysteine level was significantly higher in patient group with PEXG when compared to those of POAG and control group and that homocysteine can play a role in pathogenesis of PEXG. Xu et al.\(^\text{[21]}\) showed that PEXG was related with increased plasma homocysteine and decreased folic acid level; however, it was not related with serum vitamin B12, vitamin B6 and methylenetetrahydrofolate reductase (MTHFR) C677T genotype. In the other study, Xu et al.\(^\text{[22]}\) found that POAG is associated with elevated plasma total homocysteine levels, but not serum folic acid, vitamin B12, vitamin B6 levels, or MTHFR C677T genotype.

It was shown that increased concentration of homocysteine and hydroxyproline in humor aqueous can be related with POAG, however in parallel to this there was no significant difference between plasma levels\(^\text{[23]}\). Furthermore, it was reported that both tear and plasma homocysteine levels of PEXG patients can be increased\(^\text{[24]}\).

Turgut et al.\(^\text{[25]}\) reported that plasma homocysteine levels of patients with PEXG were higher than those of control group and that vitamin B12 level was lower. Interestingly, they showed that vitamin B6 level significantly increased in POAG and NTG patients. In conclusion, they reported that further research is necessary for the use of homocysteine and vitamin B6 as the determinants of hyperhomocysteine. Similarly, no relationship was found between NTG patients and plasma homocysteine level and it was reported that it is unlikely to have a role as a risk factor in pathogenesis of these patients\(^\text{[26]}\).

Turkeu et al.\(^\text{[27]}\) reported that the patients with PEXS plus NTG had higher plasma homocysteine levels than the patients with PEXS or PEXG and the healthy controls. The treatment of hyperhomocysteinemia by taking low-cost vitamin B12 and folic acid preparations may prevent additional vascular problems.

In our study, diet of 77 glaucoma (15 NTG, 36 POAG and 26 PEXG) and 40 healthy control patients were analyzed and serum vitamin B12 and folic acid levels were evaluated. Serum vitamin B12 level was found to be low in 16 of glaucoma patients and below normal serum level in 2 people in the control group. Folic acid level was found to be low in one individual in each group. Analysis of the diet of the patients with low vitamin B12 level showed that vitamin B12 intakes of 15 patients were deficient. Furthermore, folic acid intake of all these 16 patients was deficient, too. According to our results, vitamin B12 serum levels were significantly decreased in glaucoma patients but not folic acid. In addition, vitamin B12 intakes were deficient in patients with low vitamin B12 level (15 patients).

When the grounds of some diseases are fully explained, satisfaction of certain deficient substances externally by natural products and use of them as a therapy also gains attention. Potential nutritional therapies are important also for glaucoma. Vitamin C, vitamin B12, lipoic acid, magnesium, melatonin, CoQ10, bilberry, ginkgo, forskolin and salvia miltiorrhiza are recommended nutritional therapies for glaucoma\(^\text{[28]}\). Therefore, intake of these vitamins by glaucoma patients in their diet is considered to be important.

Glaucoma is a risk factor for the families with the history of POAG. While some of the research mentioned above reported that vitamin B12 and folic acid deficiency were not a risk factor for POAG, some indicated these two as a risk factor. In our study, vitamin B12 level was found to be low in approximately 20% of glaucoma patients; folic acid levels were found to be low only in one patient.

**Conclusion**

It was found significant statistical difference between glaucoma patients and healthy control groups regarding serum level of vitamin B12 but not folic acid, determination of low serum level of vitamin B12 in 16 of 77 glaucoma patients may indicate the importance of dietary vitamin B12 intake in glaucoma patients. Low patient number is a limitation of this study therefore; large scaled studies are needed for establishing the association of vitamin B12 and folic acid with glaucoma.

**References**

2. Ozarslan M. *Relationships between ocular pulse amplitude and age, central corneal thickness, intraocular*
pressure, axial length, anterior chamber depth, systolic-diastolic blood pressure and pulse pressure in healthy persons and in adults with glaucoma through Turkish population. Thesis of residence in Department of Ophthalmology, Ondokuz Mayis University Medical School, Samsun, Turkey, 2010.


