Vitamin B_{12} Deficiency and Thrombosis

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SUMMARY

Purpose: Thromboembolic diseases are multi-factorial. Hyperhomocysteinemia (HH) is one of these factors. Vitamin B_{12} deficiency is acquired reasons for HH. HH is a risk factor for arterial and venous thrombosis. The incidence of thrombosis is high due to increase of homocystein levels in HH associated with vitamin B12 deficiency. However, this situation is not clear and there are opposing views. In our study, we aimed to determine the frequency of thromboembolic events in individuals with vitamin B_{12} deficiency.

Methods: One hundred forty-three patients (63 female, 80 male, mean age 59±13 years), who were diagnosed as having vitamin B_{12} deficiency (below 200 pg/ml), were included in the study retrospectively. The history of arterial and venous thrombosis for a period of last three years before the time of diagnosis and was searched. The thromboembolic events, which are detected via clinical and laboratory studies and via imaging, were determined. The control group was consisted of 129 healthy subjects (62 female, 67 male; mean age 58±8) of appropriate age, sex and excluded criteria whose B_{12} level was normal.

Results: In control group, mean B_{12} level was 244±32 pg/ml. The patients' mean B_{12} level was 136±58 pg/ml. Fourteen (9.8%) thromboembolic (10 coronary artery disease, two deep venous thrombosis, two cerebrovascular) events were determined in vitamin B_{12} deficiency group, whereas this ratio was 3.9% in the control group (four coronary artery disease, one deep venous thrombosis). This ratio was not statistically significant different ($p > 0.05$).

Conclusion: Alone HH due to vitamin B_{12} deficiency did not increase the risk of thrombosis.

Key words: Arterial thrombosis, venous thrombosis vitamin B_{12} deficiency.

B_{12} Vitamini Eksikliği ve Tromboz

ÖZET

Amaç: Tromboembolik hastalıklar multi-faktöriyeldir. Hiperhomosisteinemi (HH) bu faktörlerdendir. B_{12} vitamini eksikliği HH için kazanılmış nedenlerinden biridir. HH arteriyel ve venöz tromboz için risk faktörüdür. B_{12} vitamin eksikliğine bağlı HH'de homosistein düzeylerindeki artışa bağlı olarak tromboz insidansı yüksektir. Ancak bu durum açık değildir karışık görüşler de vardır. Çalışmamızda B_{12} vitamini eksikliği saptanan kişilerde tromboembolik olay sıklığını belirlemeyi amaçladık.

Yöntem: Retrospektif olarak kazanılmış B_{12} vitamin eksikliği saptanan (200 pg/ml altında) 143 hasta (63 kadın, 80 erkek, yaş ortalaması 59±13 yıl) çalışmaya alındı. Tanı anında ve 3 yıl önceki arteriyel ve venöz tromboz öyküsü sorgulandı. Klinik, laboratuar ve görüntüleme ile saptanmış tromboembolik olaylar belirlendi. Kontrol gurubu olarak yaş ve cins gurubu uygun, B_{12} vitamin düzeyi normal, hasta gurubundaki dışlama kriterlerine uyan 129 sağlıklı kişi (62 kadın, 67 erkek, yaş ortalaması 58±8) alındı.

Bulgular: Kontrol gurubunda ortalama B_{12} düzeyi 244±32 pg/ml, olgularımızda ise 136±58pg/ml idi. B_{12} vitamini eksikliği olan olgularda %9.8 oranında tromboembolik (on koroner arter hastalığı, iki derin venöz trombozu, iki serebrovasküler) olay saptanır iken kontrol gurubunda bu oran %3.9 (dört koroner arter hastalığı, bir derin venöz tromboz) idi. Bu oran istatistiksel olarak anlamlı değildi ($p>0.05$).

Sonuç: B_{12} vitamin eksikliği nedeni ile oluşan HH tek başına tromboz riskini arttırmamaktadır.

Anahtar kelimeler: B_{12} vitamin eksikliği, arteriyel tromboz, venöz tromboz

INTRODUCTION

Thromboembolic diseases are multi-factorial. Hyperhomocysteinemia (HH) is one of these factors (1). HH can be acquired or hereditary. B_{12}, B_6 vitamins, folate deficiency, renal insufficiency, hypothyroidism, type 2 diabetes, inflammatory bowel disease, climacterium, old age, solid tumors (especially ovary, breast, pancreas), acute lymphoblastic leukemia and medicines (niacin, fibrates, isoniazides, L-dopa, teophilin, metformine, methotrexate, trimethoprim, fenitoin etc.) are acquired reasons for HH. It is observed at different degrees in enzyme mutations in the metabolic pathway of homocystein genetically and especially in methylene tetrahydropholate reductase gene (MTHFR) mutations (2,3).

HH is a risk factor for arterial and venous trombosis. It is asserted that in deficiency of vitamin B_{12} thrombotic incidences increase due to increase in homocystein and upon substituting the vitamin this risk decreases (4-7). However, this situation is not clear and there are opposing views (8). Body stores for cobalamin are of the order of 2-3 mg, sufficient for 3-4 years if supplies are completely cut off (2).

In our study, we aimed to determine the frequency of thrombotic incidences in the individuals who were diagnosed as having vitamin B_{12} deficiency.

MATERIALS AND METHODS

Between 1999 and 2005, 143 patients with anemia (63 female, 80 male, mean age 59±13), who were diagnosed as having vitamin B_{12} deficiency (below 200 pg/ml) were included in the study retrospectively. We took approval for this research from the ethical committee of Adnan Menderes University Medical Faculty.

Those with renal deficiency or pregnancy, multiple myeloma, known B_{12} and B_6 deficiency, thyroid disease, myelodysplastic syndrome, folat deficiency, iron deficiency anemia, and using medicine which are known to affect homocystein and B_{12} metabolism, were excluded from the study.

The history of arterial and venous thrombosis for a period of last three years before the time of diagnosis was searched. The thromboembolic events, which are detected using clinical, laboratory and imaging studies, were determined. The healthy control group, who admitted for checkup, was consisted of 129 persons (62 female, 67 male; and mean age 58±8 years) with appropriate age, sex, excluded criteria, and normal vitamin B_{12} level. Risk factors for arterial and venous thrombosis were similar in control groups.

B_{12} was studied with turbidimetric method (Beckman coulter access immunoassay system). Blood specimens for hematological parameters were collected in tubes with EDTA and were analyzed after 1ï 2 h. These tests were determined with Coulter Counter Gen-S.

The statistical analysis was carried out by using Statistical Package for Social Sciences (SPSS), version 13.0. Data were presented as mean ± standard deviation (S.D.). For the comparisons of parametric and non-parametric results, student-t test and chi-square tests, respectively were used. Significance was accepted as $p < 0.05$.

RESULTS

In the patient group, mean vitamin B_{12} level was 136±58 pg/ml. Fourteen (10 male, 4 female) (9.8) thromboembolic (9 coronary artery disease, 2 deep venous thrombosis, 2 cerebrovascular) events were determined in patient group, whereas this ratio was 3.9% (4 female, 1 male) in the control group. These events were 4 coronary artery disease and 1 deep venous thrombosis. This difference for total group was not found statistically significant ($p>0.05$).

Table 1: Demographic and laboratory features of patient and control group

	Patients (n=143)	Control (n=129)	p
Sex (male/female)	80/63	67/62	>0.05
Mean age (year)	59±13	58±8	>0.05
Hemoglobin (gr/dl)	10±3.1	13±2.9	<0.05
Mean corpuscular volume (f/l)	102±11	88±7	<0.05
B12 level (pg/ml)	136±58	244±32	<0.05
Homocystein (μmol/L)	39±17(n=36)	-	-
Thrombosis			
Male (n)	10	1	**0.003**
Female (n)	4	4	>0.05
Total (n)	14	5	>0.05

This ratio was not statistically significant different (p> 0.05). But trombosis rate for male groups was found as statistically significant (p= 0.003). In table 1, the features of the patiens and the results of thrombosis are shown. Homocystein (H) levels could be examined in only 36 patients. And these levels were high more than 15 micromoles/L in all of them (2).

DISCUSSION

In this study, the frequency of thromboembolic events in the patients with vitamin B_{12} deficiency was not significantly higher than control group, although this frequency was higher than two folds. Moreover thromboembolic events in male patients were more frequently seen as statistically significant as male controls.

HH due to hereditary homocystinuria is a risk factor myocardial infarction, peripheral artery disease, carotid artery stenosis, and recurrent low extremity venous thrombosis (2). How HH causes thrombosis is not clear. It is considered that HH causes endothelial dysfunction in animal models and especially in small veins. Desquamation of the endothelium cells, lipoprotein oxidation with low density, increase in adhesion of monocyte to endothelium, activation of factor 5, increased thrombin activation, damaged fibrinolytic potential, vascular inflammation, and apoptosis are some of the proposed mechanisms (9,10).

Remacha et al. (7) found vitamin B_{12} deficiency as a risk factor especially in arterial thrombosis. However, they did not examine the other causes and risk factors of thrombosis such as overweight, smoking, family history, cholesterol levels, early menopause, high blood pressure, and diabetes. We also did not search them.

In case control studies, Perry et al. (11) have determined high H as an independent risk factor for stroke. In a meta-analysis, although high H level is an independent risk factor ischemic heart disease, whether vitamin support would decrease cardiovascular risk, is not clear. But genetic evaluation could not be evaluated in prospective studies (12). In a study by Oger et al. (13) moderate level of H and low level of vitamin B_{12} have been considered to be risk for venous thromboembolism (VTE). However, the reasons, acquired or hereditary, that caused atherothrombosis, were not evaluated. In this study, the average age is 67±4 years. It is known that vitamin B_{12} deficiency increases with age. It was emphasized that the relation among H level, vitamin B_{12} and thrombosis should be evaluated in young patients with VTE in this study.

Hereditary thrombophilia may be caused by reasons such as factor V leiden, prothrombin G20210A gene, MTHFR C677T mutation. In healthy individuals while mutation of factor V leiden was 4.8.%, of prothrombin G20210T gene was 2.7%, of MTHFR was 10-20%, in presence of thrombosis they were 40%, 16%, and 1-20%, respectively (14). While high level of H is a risk factor especially for peripheral artery disease, MTHFR gene mutation is frequently detected in these patients (15).

In this study which HH was evaluated in venous deficiency, attention was called to mutation MTHFR gene (16).

In an evaluation made by Ridker et al (17) they have emphasized that besides high levels of H, reason for hereditary thrombophilias should also be considered in patients with VTE. In a study, which considers that in patients with HH and VTE pernicious

anemia can occur as the first finding; pernicious anemia was not found to be high. However, it was emphasized that it should be considered after other thrombophilia reasons are excluded (18).

In a study by Nygard et al. (19) they have evaluated the relation between cardiovascular mortality risk and H level in 7991 male 8585 female, they found out that H level increased with age and it was higher in males and in those smoke. They have emphasized that together with smoking, old age, male sex, high blood pressure, increased cholesterol values, HH is also important

On the contrary, in a study, 80 patients with VTE were compared with 123 healthy controls and it was found out that while H level is affected by age, sex and B_{12} level, it was not found as a risk factor for VTE (8). Differences between the results of a study carried out in Far East and our study may be indicative of differences among races.

In another presentation of case, deep venous thrombosis was developed under treatment due to vitamin B_{12} deficiency, but there was mutation of factor V leiden in this patient . Attention was drawn to more than one etiology for thromboembolic diseases (20). In case with pernicious anemia of Marie et al. (21) H was high. Mesenteric venous thrombosis was developed in the case. However, there was also prothrombin G20210T mutation which is a clear risk factor for thrombosis in the case. In three young cases (their ages were 24, 35, and 30, respectively), venous thrombosis and HH as a result of vitamin B_{12} deficiency were interrelated to each other. However, evaluation was not performed for hereditary thrombophilias (22,23). High level of H is the most sensitive marker of clinical cobalamine deficiency. Methylmalonic acid is at least as sensitive as H and it is more specific. In clinical cobalamine deficiency, vitamin B_{12} level is usually below 148 pg/ml. In 96% of these cases, H increased. In old people, subclinical cobalamine deficiency was observed at a ratio of 10% (below 200pg/ml). In this situation, mild increase in H is observed, too (2). In our cases the average vitamin B_{12} level was 136±58 pg/ml. H levels could be examined in only 36 patients. And these levels were high more than 15 micromoles/L in all of them.

In a review by Den Heijer (24), he commented on the relation between vitamins and thrombosis. Moreover he claimed a relation between low vitamin B level and thrombosis, and put emphasis especially on their statistical restrictions and contradictions. During acute thrombotic incidence, H indicates that renal deficiency, vitamin deficiency (besides B_{12}, folate, vitamin B_6), smoking, insufficient physical training, high blood pressure can be different in males and post-menopausal women. It is emphasized that the relation which can be established between HH and thrombosis in some diseases, can be deceptive and that differences in the enzymes in the H metabolism can be important. This situation can affect the results of the study (25). In order to decrease the cardiovascular risk with regard to public health, the role of providing folate or vitamin B_{12} should also be discussed (1-3,5,24). In a study, males older than 70 years old, it was interpreted that the low level of vitamin B_{12} constitutes a risk with respect to VTE. In these patients, there was HH. The frequency of mutation of factor V leiden also was found to be increased. The analysis of the mutation of the MTHFR gene was obscure (26).

While ten coronary artery diseases, two deep venous thrombosis, two cerebrovascular events were determined in our patients with B_{12} deficiency, there were four coronary artery diseases, one deep venous thrombosis events were observed in the control group. We could not evaluate the reasons, which can cause hereditary thrombophilias. As in the current study, coronary artery disease increases with age (28). In the meta-analysis of Den Heijer et al (27), it was concluded that the result of high H related to MTHFR gene mutation is a risk factor for VTE and vitamin B_{12} deficiency is a common problem in elderly subjects (26,28).

In conclusion, Alone HH due to vitamin B_{12} deficiency did not increase the risk of thrombosis. The larger prospective and randomized studies needs in the detection of risks for thrombosis in vitamin B_{12} deficiency.

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