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RESEARCH ARTICLE

The Relationship between Vitamin B12 Deficiency and Red Cell Distribution Width-Platelet Ratio

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ABSTRACT

Objective: We aimed to investigate the relationship between vitamin B12 deficiency and red cell distribution width-platelet ratio (RPR), and the variations in the parameters on vitamin B12 treatment.

Methods: One hundred fifty-four patients with untreated vitamin B12 deficiency (56% men, mean age: 50 ± 12.7 years) (untreated group), 86 patients with vitamin B12 deficiency (62% men, mean age: 42 ± 20.7 years) on vitamin B12 treatment (treated group), and 92 age- and sex-matched control group (54% men, mean age: 45 ± 15.1 years) were included in the study. Hematological parameters were evaluated by the method of laser-based flow cytometric impedance, using an automated blood cell counter (ABX Pentra 120 Hematology Blood Analyzer).

Results: RPR was significantly reduced in treated group compared with untreated group (4.88 ± 1.06; 6.13 ± 1.27; $p<0.001$, respectively). RPR, red cell distribution width (RDW) and mean platelet volume (MPV) were significantly different in untreated group compared to controls ($p<0.05$). There were no differences in platelet count in the three groups ($p>0.05$).

Conclusion: We proposed that vitamin B12 deficiency has effects on RPR and supplementation with vitamin B12 corrects the RPR levels. *J Clin Exp Invest 2016; 7 (3): 211-215*

Key words: Vitamin B12 deficiency, red cell distribution width-platelet ratio, vitamin B12 treatment, Mean platelet volume

Vitamin B12 Eksikliği ve Eritrosit Dağılım Genişliği-Trombosit Oranıyla İlişkisi

ÖZET

Amaç: Biz B12 vitamini eksikliği ve eritrosit dağılım genişliği-trombosit oranı (RPR) arasındaki ilişkiyi ve vitamin B12 tedavisinin bu parametrelerdeki değişimini araştırmayı amaçladık.

Yöntemler: Çalışmaya 154 tedavi olmamış vitamin B12 hastası (%56'sı erkek, ort. yaş: 50 ± 12,7) (tedavi olmayan grup), 86 vitamin B12 eksikliği olan ve tedavi alan hastalar (%62'si erkek, ortalama yaş: 42 ± 20,7) (tedavi alan grup) ve 92 yaş ve cinsiyet uyumlu kontrol grubu (%54'ü erkek, ort. yaş: 45 ± 15,1 yıl) dahil edildi. Hematolojik parametreler, lazer tabanlı akım sitometri empedansı yöntemiyle otomatik kan hücre sayıcıyla (ABX Pentra 120 Hematology Blood Analyzer) ölçüldü.

Bulgular: RPR, tedavi alan grupta tedavi almayan gruba göre anlamlı olarak düşük tespit edildi (4,88 ± 1,06; 6,13 ± 1,27; $p<0.001$, sırasıyla). RPR, eritrosit dağılım genişliği ve ortalama trombosit hacmi tedavi almayan grupta kontrol grubuyla karşılaştırıldığında anlamlı fark vardı ($p<0.05$). Üç grup arasında, trombosit sayılarıyla anlamlı fark yoktu ($p>0.05$).

Sonuç: Vitamin B12 eksikliğinin RPR üzerinde etkisi olup, vitamin B12 eksikliğin tedavisiyle RPR seviyelerinin düzeldiğini önermekteyiz.

Anahtar kelimeler: Vitamin B12 eksikliği, eritrosit dağılım genişliği-trombosit oranı, B12 vitamini tedavisi, Ortalama trombosit hacmi

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INTRODUCTION

The degradation in methionine metabolism, the mitochondrial methylmalonic acid-coenzyme B12 vitamin pathway in the mitochondria, and DNA synthesis results in replaced concentrations of intermediary metabolites and are also accountable for variations in some hematological parameters in patients with vitamin B12 deficiency [1-3]. Eventually, the patients with vitamin B12 deficiency will have clinical symptoms of macrocytic anemia and hyperhomocysteinemia. The high level of homocystein results in cardiovascular diseases. Numerous studies have suggested that endothelial dysfunction is strongly linked to hyperhomocysteinemia [4-6]. Also, vitamin B12 deficiency leads to a lack of methylcobalamin, which has been related to significant neurological diseases [7]. The complete blood count is one of the most commonly used easy tests in clinical practice and also has been shown in several studies to predict various disease outcomes and mortality. The red cell distribution width (RDW) reflects erythrocyte anisocytosis and is related to inflammation. Activated red blood cells may have a crucial role in inflammation. Mean platelet volume (MPV) is a hemogram parameter that authors mentioned in association with inflammation [8,9]. Moreover, cardiovascular diseases are characterized by subclinical inflammation. A novel, inexpensive, and easily calculated index, red cell distribution width-platelet ratio (RPR) is used in clinical practice for predicting mortality in patients with chronic diseases [10]. RPR is currently studied as an inflammatory marker and predictor of several cardiovascular diseases.

Our current data regarding the association between RPR and vitamin B12 deficiency is not yet determined. We aimed to evaluate the relationship between RPR levels and vitamin B12 deficiency as well as effects of vitamin B12 treatment.

METHODS

This study included 154 patients with untreated vitamin B12 deficiency (56% men, mean age: 50 ± 12.7 years), 86 patients vitamin B12 deficiency (62% men, mean age: 42 ± 20.7 years) on vitamin B12 treatment, and 92 age- and sex-matched healthy individuals (control) (54% men, mean age: 45 ± 15.1 years). Exclusion criteria for patients were as follows: the presence of folate and iron deficiency anemia or chronic diseases (such as liver diseases, renal diseases, diabetes mellitus, hypertension, dyslipidemia, cardiovascular diseases and thyroid diseases). The study complied with the Helsinki Declaration and was approved by the local ethics committee of the Muğla Sıtkı Kocman University School of Medicine, Muğla, Turkey. The patients who underwent vitamin B12 evaluation were grouped based on the serum concentration of vitamin B12. The vitamin B12 deficiency was defined as low serum vitamin B12 concentration (≤ 197 pg/ml). The levels of vitamin B12 in vitamin B12-deficient patients were 102 ± 28 pg/ml (mean ± SD; females, 112 ± 37 pg/ml and males, 91 ± 38 pg/ml). The lowest level of vitamin B12 measured in the patients was 65 pmol/l and the highest level was 189.6 pg/ml. The reference range of serum concentration of vitamin B12 was 197-771 pg/ml in the laboratories of our institution. The complete blood count analyses were performed using an automated blood cell counter (ABX Pentra 120 Hematology Blood Analyzer). The serum concentration of vitamin B12 was evaluated by electrochemiluminescent assay performed in the detection of Roche diagnostics, Cobas, 6000. Patients' and controls' characteristics and laboratory data, white blood cell count (WBC), neutrophil count, lymphocyte count, hemoglobin (Hb) count, mean corpuscular volume (MCV), red cell distribution width (RDW), platelet, MPV, platelet distribution width (PDW) and RPR were detected.

Statistical Analysis

SPSS software (SPSS 20.0 software program IBM Corp, Armonk, New York, USA) used for statistical analysis. Continuous variables were described as mean ± standard deviation. The Mann-Whitney U-test and Student's t-test were used for determining the differences in patients' characteristics and laboratory data of the groups. Categorical data were compared with chi-square test. Statistical significance was accepted at $p<0.05$ level.

RESULTS

The age and gender of participants were not significantly different between three groups (p=0.63, p=0.74, respectively) (Table 1). There were no significant differences between the study groups in terms of following hematological parameters: WBC and platelet count (all $p>0.05$). Hemoglobin value was significantly different in untreated group compared with treated group (p=0.004). MCV was significantly different in untreated group compared with control group (p=0.031). RDW was significantly higher in untreated group compared with control group (15.6 ± 1.8; 13.1 ± 0.4; respectively, p=0.012) (Table 1). Significantly

increased MPV values were found in untreated group compared to the control group (9.6 ± 1.12; 7.9 ± 0.7, p=0.034, respectively) (Table 1). MPV and RDW were not significantly different in untreated group compared to the treated group (p>0.05). RPR was significantly higher in patients with untreated group compared to the patients with treated group (6.93 ± 1.27; 4.59 ± 1.02, respectively) (p<0.001) (Table 1).

In addition, when we performed a correlation analysis, it indicated a positive correlation between vitamin B12 and RPR (r=0.316, p<0.001) and WBC (r=0.245, p=0.001) and a negative correlation with age (r=-0.165, p=0.001) and Hb (p=-0.114, p=0.146) (Table 2). However, the levels of vitamin B12 were not correlated with any other hematological parameters (p>0.05).

Table 1. The main characteristics and hematological parameters for the three groups

Variables	Vitamin B12 deficiency Group	Vitamin B12 treated Group	Control Group
Age (year)	50.2±12.7	43.7±20.7	45±15.1
Male gender, (%)	56	62	54
WBC (x10^9L)	5.8±3.1	5.32±3.5	4.32±2.3
Hb (g/dl)	11.8±2.4[a]	13.7±1.3	14.9±3.6
MCV (fl)	93.3±9.7[a]	84.4±6.1	86.4±7.6
Platelet count (10^3/µl)	273.5±81.32	254.2±74.8	249.8±92.4
RDW	15.6±1.8[a]	14.2±0.7	13.1±0.4
MPV (fl)	9.6±1.12[a]	8.4±1.28	7.9±0.7
RPR	6.93±1.27[a,b]	4.59±1.02	4.88±1.85
Vitamin B12(pg/ml)	102.7±28.8[a,b]	1208±607.4	598.2±311.5

WBC: White blood cell; Hb: Hemoglobin; MCV: Mean cell volume; MPV: Mean platelet volume; RDW: Red cell distribution width; RPR: Red cell distribution width platelet ratio

[a] p<0.05 versus control group; [b] p<0.05 versus vitamin B12 treatment

Table 2. Correlation coefficients between vitamin B12 and other parameters

	(r)	p value
Age	0.165	0.001
Hb	0.114	0.146
WBC	0.245	0.001
RPR	0.316	<0.001

Hb: Hemoglobin; WBC: White blood cell; RPR: Red cell distribution width platelet ratio

DISCUSSION

We mainly found that RPR was significantly decreased in patients with vitamin B12 deficiency compared to patients receiving vitamin B12 treatment. The RPR projects the severity of inflammation. RPR could be determined to predict the mortality of some chronic diseases as a strong and crucial marker. The MCV levels may improved in late stages in patients with vitamin B12 deficiency after vitamin B12 treatment [11,12]. In our study, vitamin B12 deficiency should potentialize inflammation conditions via RPR levels. In addition, MPV, RPR and RDW were significantly different in patients with vitamin B12 deficiency compared to control groups. MPV reflects the extent of circulating platelets and RDW reflects the extent variability of erythrocytes [13]. Various studies have indicated that there is a relationship between the two hemogram parameters and overt or subclinical inflammatory conditions [14-16]. It is well known that the level of homocysteine in serum increased in patients with vitamin B12 deficiency. The increasing homocysteine pathway leads to inflammation in patients with vitamin B12 deficiency [17].

The results of the current study support the hypothesis. The increased level of serum homocysteine is a risk factor for cardiovascular diseases [18], and MPV and RPR have been investigated as a link to cardiovascular diseases as well. Therefore, we may propose that homocysteine; RPR and MPV are involved in the course of the inflammation. Similarly, it was indicated that the homocysteine promoted platelet activation [17,18]. A deranged growth and disrupted

division of hematopoietic cells consist seen in vitamin B12 deficiency leads to defects in DNA synthesis. RDW indicates heterogeneity and equivalent of anisocytosis of red blood cell. It was documented that high levels of RDW predicts elevated red blood cell destruction in iron, folate, and vitamin B12 deficiency [19-21]. Eventually, different-sized erythrocytes may be expressed and elevated RDW may result in increased RPR. In other ways, the inflammation presenting in vitamin B12 deficiency may lead to elevation of the parameters as following RDW, MPV and RPR. It was indicated that vitamin B12 deficiency is related to inflammation via serum hyperhomocysteine and high level of RDW has been reported in literature [22]. We determined that MPV levels are increased in patients with vitamin B12 deficiency compared to healthy individuals. In a study, MPV was found to be lower in patients with vitamin B12 deficiency than in the patients without vitamin B12 deficiency as a result of production of smaller platelets [23,24]. We support that vitamin B12-deficient patients have inflammatory conditions and MPV reflects the inflammatory position. Similarly, MPV was found to have a relationship with some inflammatory processes such as coronary ischemia, preeclampsia, ischemic stroke, rheumatoid arthritis, and inflammatory bowel disease [25-30]. There may be various limitations in our study. First, these findings should be explained by larger samples. In addition, we should measure the serum level of homocysteine in the participants.

In conclusion, we think that RPR should be an indicator of vitamin B12 deficiency and the parameter may be ameliorated in patients under vitamin B12 treatment except for other hematological parameters.

Declaration of Conflicting Interests: The authors declare that they have no conflict of interest.

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