

B Group Vitamin Levels in Patients With Chronic Obstructive Pulmonary Disease and The Relation Between Pulmonary Functions

Kronik Obstrüktif Akciğer Hastalığı Olan Olgularda B Grup Vitamin Düzeyleri ve Solunum Fonksiyonları İle İlişkisi

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ABSTRACT

Introduction: We aimed to evaluate the B group vitamin levels in patients with COPD and to determine the relationship between vitamin levels and pulmonary functions.

Material and Method: Fifty four patients with COPD, who were initially diagnosed during the admission to the outpatient clinic, were studied. Pulmonary function tests and assays of vitamins of COPD patients, healthy subjects were evaluated.

Results: All the B group vitamin levels were lower in the COPD group. B1, B2, B12 vitamin levels correlated with smoking (pack/year) ($r=-0.33$, $r=0.265$, $r=0.483$, respectively, $p<0.05$ for all values), forced expiratory volume in one second (FEV1) ($r=0.372$, $r=0.474$, $r=0.777$ respectively, $p<0.05$ for all values), and FEV1/forced vital capacity (FVC) ($r=0.418$, $r=0.430$, $r=0.787$ respectively, $p<0.05$ for all values). B6 vitamin levels correlated with FEV1/FVC values ($r=0.290$, $p<0.05$).

Conclusion: This study confirms a decreased level of B group vitamins in patients with COPD. It may be suggested that B group vitamin supplementation may have a beneficial effect on oxidant/antioxidant imbalance in such patients. Future work is needed to explore the possible relationship between the intake of B group vitamins, systemic and local pulmonary assessment of antioxidant capacity/antioxidant molecules, and lung function. (*Tur Toraks Der 2008;9:88-92*)

Key words: B group vitamins, COPD, antioxidants, pulmonary functions

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ÖZET

Giriş: KOAH' lı olgularda B grup vitamin düzeylerini ölçmeyi ve vitamin seviyeleri ile solunum fonksiyonları arasındaki ilişkiyi araştırmayı amaçladık.

Gereç ve Yöntem: Göğüs hastalıkları polikliniğine başvuran 54 KOAH' lı olgu çalışmaya alındı. KOAH' lı olguların ve sağlıklı kontrol grubun solunum fonksiyon testleri ve vitamin düzeyleri ölçüldü.

Bulgular: KOAH' lı olgularda tüm vitamin seviyeleri düşük izlendi. B1, B2, B12 vitamin seviyeleri sigara (paket/yıl) ile (sırasıyla, $r=-0.33$, $r=0.265$, $r=0.483$, tümü için $p<0.05$) FEV1 ile (sırasıyla, $r=0.372$, $r=0.474$, $r=0.777$ için $p<0.05$) ve FEV1 / FVC değeri ile (sırasıyla, $r=0.418$, $r=0.430$, $r=0.787$, tümü için $p<0.05$) korelasyon gösterdi. B6 vitamin seviyeleri yalnızca FEV1 / FVC değeri ile korelasyon gösterdi ($r=0.290$, $p<0.05$).

Sonuç: Bu çalışma KOAH' lı olgularda B grup vitamin seviyelerinin azaldığını gösterdi. Bu grup hastalarda B grup vitamin desteğinin oksidan/antioksidan denge üzerine faydalı olacağı düşünülebilir. B grup vitamin alımı ile sistemik ve lokal antioksidan kapasite, antioksidan moleküller ve akciğer fonksiyonları arasındaki olası ilişkiyi göstermek için ileri çalışmalara ihtiyaç vardır. (*Tur Toraks Der 2008;9:88-92*)

Anahtar sözcükler: B grup vitaminler, KOAH, antioksidanlar, solunum fonksiyonları

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INTRODUCTION

Chronic Obstructive Pulmonary Disease (COPD) is a condition characterized by progressive and largely irreversible airway obstruction and influx of inflammatory cells into the lungs [1]. While the precise mechanisms of the pathogenesis of COPD have not been defined, increased oxidant burden and protease-antiprotease imbalance have been proposed. The increased oxidant burden derives from the fact that cigarette smoke, the main etiological factor in the pathogenesis of COPD,

contains an estimated 10^{14} radicals/puff and about 4700 chemicals that include peroxynitrite, superoxide radical and oxides of nitrogen. The oxidant burden in the lungs is further enhanced in smokers by the release of reactive oxygen species (ROS) from alveolar macrophages and sequestered neutrophils in the lung [2].

There are several lines of evidence that suggest an imbalance between oxidants and antioxidants in the lung and blood in smokers and patients with COPD [3-5]. Most investigations have centered on a few parameters

of the oxidant-antioxidant balance, such as decreased plasma protein sulfhydryl groups, decreased total antioxidant capacity, increased lipid peroxidation products [3], increased superoxide generation from peripheral neutrophils [6] and increased F2-isoprostanes [7].

Epidemiological studies have shown that some of the markers of oxidative stress may be associated with reduced lung function. An example of this is the inverse relationship between circulating neutrophil numbers and the FEV₁ [8]. In addition, a high dietary intake of the antioxidant vitamins C and E has been shown to relate to better lung function in the general population [9] and to a lower prevalence of chronic bronchitis in smokers [10], suggesting the protective effect of these vitamins.

It is known that B group vitamins also have an antioxidant capacity [11]. To our knowledge there is no study which evaluates this vitamin levels in patients with COPD. The primary outcome measure of this study was to evaluate the B group vitamin levels in patients with COPD and the secondary outcome measure was to determine the relationship between vitamin levels and pulmonary functions.

MATERIAL AND METHOD

The Ethics Committee of Firat University Medical Faculty reviewed and approved the protocol, and all patients signed informed consent after the nature of the study had been fully explained prior to their participation.

Patients: Fifty four patients with COPD, (group 1) who were initially diagnosed on admission to the outpatient clinic, were studied. The diagnosis of COPD was made by a respiratory physician on the basis of current or ex-smoking and largely irreversible airways obstruction, with <15% improvement in baseline forced expiratory volume in one second (FEV₁) in response to an inhaled β_2 agonist [12]. The clinical condition of the patients was stable, with no acute exacerbations of COPD in the month prior to entry into the study. Patients had no other concurrent pulmonary or systemic disease or evidence of any upper or lower respiratory tract infection. All COPD patients were male and current smokers. Information regarding the duration and frequency of nicotine-smoking was obtained from the participants. The pack/year score was calculated as follows: the number of cigarettes smoked per day, divided by 20, multiplied by the number of years during which the individual had smoked.

All COPD patients were treated with a standardized therapy including inhaled salmeterol/formoterol, inhaled tiotropium bromide, oral theophylline and inhaled fluticasone/budesonide after taking blood samples for assaying vitamin levels.

Control subjects: Thirty healthy male subjects recruited from hospital staff and the general population participated in the study. Fifteen subjects (group 2) were current smokers, 15 subjects (group 3) had never been smokers. None were receiving any medications. None had a history of lung disease, other concurrent pulmonary or systemic disease.

Spirometry: The respiratory function tests were performed using a spirometry device (Superspiro, Micromedical Limited, Rochester, England). Maximal Expiratory Flow Volume curves were obtained. Three acceptable and at least two reproducible curves were obtained in each subject. The highest value of FEV₁ was selected for analysis.

Assay of B group vitamin levels: Following an overnight fast of 12 h, blood samples were taken for vitamins B1, B2, B6, and B12. Serum vitamin B12 levels were measured by IMMULITE 2000 Analyzer using a Chemiluminescence kit (DPC, diagnostic Products Corporation, Los Angeles, CA). Vitamin B6 was measured as pyridoxal-5-phosphate. Serum vitamin B1, B2, and B6 levels were measured by an HPLC method (Recipe Chemicals Instruments GmbH, Germany) on a heparin plasma sample.

Statistical Analysis: Data were analyzed using the statistical package for the Social Sciences (SPSS) v12.0. Results were given as group means \pm standard deviations (SD). A p value of <0.05 was considered statistically significant. Analysis of variance (ANOVA) was used to evaluate the difference among the three groups. After applying ANOVA, post-hoc comparisons were carried out. Correlations were evaluated by Pearson's test.

RESULTS

Demographic characteristics, pulmonary function test (PFT) parameters and B group vitamin levels of all participants are shown on Table 1. There was no difference between groups in age and sex distribution.

When COPD groups and both controls were compared according to B group vitamin levels, it was seen that all B group vitamin levels were lower in the COPD group and, except B6 vitamin, all vitamins showed a statistical significance. Mean difference, standard error, levels of statistical significance in post-hoc comparisons between the 3 groups, using the Bonferroni t-test are shown in Table 2.

Table 1. Demographic characteristics, PFT parameters, B group vitamin levels of all participants

	COPD group (Group 1, n=54)	Control group Current smoker (Group 2, n=15)	Control group Never smoker (Group 3, n=15)
Age	68.92 \pm 6.59	65.46 \pm 5.18	67.40 \pm 4.61
Smoking(pack/year)	33.98 \pm 11.62	30.13 \pm 9.42	0†
FEV ₁ (%predicted)	39.22 \pm 13.50*	82.86 \pm 4.61	84.26 \pm 5.21†
FEV ₁ /FVC (%)	63.81 \pm 9.05*	87.80 \pm 3.85	89.80 \pm 3.80†
Vitamin B1 (μ g/l)	31.66 \pm 9.15	36.14 \pm 6.19	39.27 \pm 6.26†
Vitamin B2 (μ g/l)	82.94 \pm 19.97*	102.43 \pm 9.40	100.20 \pm 4.13†
Vitamin B6 (μ g/l)	18.20 \pm 8.12	21.46 \pm 3.04	21.74 \pm 2.18
Vitamin B12(pg/ml)	314.68 \pm 28.73*	406.00 \pm 12.84	412.53 \pm 22.60†

Data are presented as mean \pm SD

*p<0.001, COPD group vs current smoker control group

† p<0.001, COPD group vs never smoker control group

When we analysed the correlations, we saw that B1, B2, and B12 vitamin levels correlated significantly with smoking (pack/year) ($r=0.33$, $r=0.265$, $r=0.483$, respectively, $p<0.05$ for all values), FEV₁ ($r=0.372$, $r=0.474$, $r=0.777$ respectively, $p<0.05$ for all values) and FEV₁/FVC ($r=0.418$, $r=0.430$, $r=0.787$ respectively, $p<0.05$ for all values). B6 vitamin levels only correlated with FEV₁/FVC value ($r=0.290$, $p<0.05$). Correlations between B group vitamin levels and FEV₁, FEV₁/FVC values are shown in fig. 1-2.

DISCUSSION

This study shows that B group vitamin levels are decreased in COPD patients when compared to both

smoker/non smoker control groups, and they are closely related with pulmonary function abnormalities. This is an expected finding, because it is known that plasma antioxidant capacity decreases in all patients with COPD, and in healthy smokers [13].

A low antioxidant capacity in plasma suggests an increased oxidant burden in the blood. Previous investigators reported increased levels of superoxide anion release from circulating neutrophils [3] and increased lipid peroxidation products in the plasma [14] of smokers and patients with COPD, supporting the concept of systemic oxidative stress in these conditions.

Smoking is known to produce systemic oxidative stress [15]. Morrison et al. showed that acute smokers have low plasma antioxidant defences compared with chronic smokers [2]. It has been reported that smoking is associated with a depletion of vitamins C and E in the blood [14]. The lower plasma trolox equivalent antioxidant capacity (TEAC) in patients with COPD and in chronic smokers could therefore result from depletion of

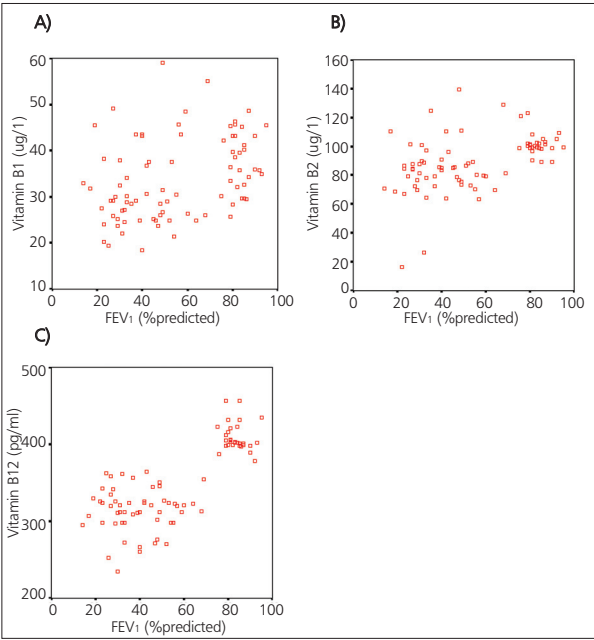


Figure 1. Correlation between FEV₁ (% predicted) and Vitamin B1 (A), B2 (B), B12 (C) levels

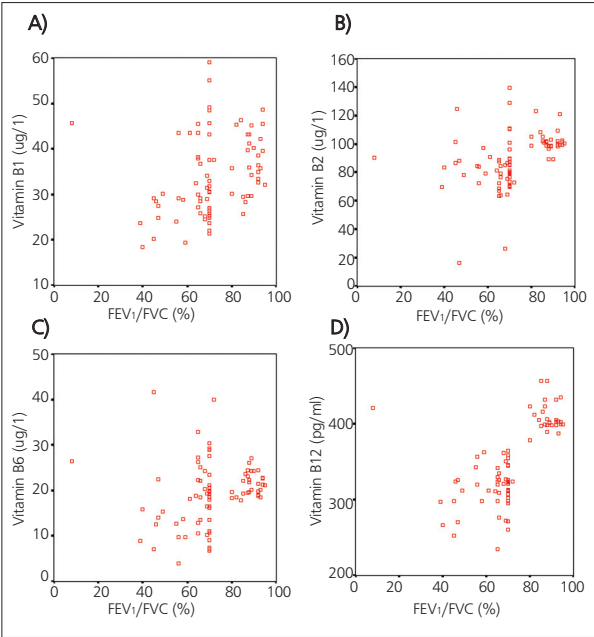


Figure 2. Correlation between FEV₁/FVC (%) and vitamin B1 (A), B2 (B), B6 (C), B12 (D) levels ($r=0.418$, $r=0.430$, $r=0.290$, $r=0.787$, $p<0.05$)

Table 2. Mean difference, standard error, levels of statistical significance in post-hoc comparisons between the 3 groups, using the Bonferroni t-test

	Group 1 vs group 2	Group1 vs group 3	Group 2 vs group 3
FEV₁(%predicted)			
Mean difference	-43,64*	-45,04*	-1,4
Standard error	3,29	3,29	4,12
P value	0.00	0.00	1.00
FEV₁/FVC (%)			
Mean difference	-23,98*	-25,98*	-2,0
Standard error	2,23	2,23	2,79
P value	0.00	0.00	1.00
Vitamin B1 (ug/l)			
Mean difference	-4,47	-7,60*	-3,13
Standard error	2,41	2,41	3,01
P value	0.20	0.007	0.90
Vitamin B2 (ug/l)			
Mean difference	-19,49*	-17,26*	2,22
Standard error	4,87	4,87	6,10
P value	0.00	0.00	1.00
Vitamin B6 (ug/l)			
Mean difference	-3,26	-3,54	-0,28
Standard error	1,97	1,97	2,46
P value	0.30	0.22	1.00
Vitamin B12(pg/ml)			
Mean difference	-91,31*	-97,84*	-6,53
Standard error	7,48	7,48	9,35
P value	0.00	0.00	1.00
Smoking(pack/year)			
Mean difference	3,84	33,98*	30,13*
Standard error	2,97	2,97	3,71
P value	0.59	0.00	0.00

*The mean difference is significant at the .05 level

such antioxidant molecules in addition to depletion of protein thiols. Northrop-Clewes et al. [16] performed a study to determine whether effects on the biochemical markers of micronutrient status were due to differences in dietary intakes between smokers and non-smokers or to the consequences of inflammatory changes caused by the oxidative stress of smoking. They concluded that serum concentrations of vitamin A, folate, and vitamin B12 and B6 markers do not appear to be influenced by smoking, although there is some influence of dietary intake on the concentrations of these nutrients in the body. In contrast, another study showed that smoking is associated with a diminished status of B2, B12 vitamins [17]. In our study we observed a significant inverse correlation between B group vitamins and smoking.

B group vitamins have antioxidant effects, but few studies have been performed to show their antioxidant capacity. Ullegaddi et al. [18] showed the antioxidant effect of B group vitamins in patients with ischemic stroke. They have reported that antioxidants supplementation with B-group vitamins enhances the antioxidant capacity, mitigates oxidative damage, and may have an anti-inflammatory effect immediately postinfarct in stroke disease. In another study, it is reported that B group vitamins have both antioxidant and prooxidant effects on lipid peroxidation under different experimental conditions [11].

In our study, we have shown that B group vitamin levels decrease, as do other antioxidant vitamins in COPD patients. The second aim of our study was to determine the relation between the levels of vitamins and pulmonary function parameters, and we saw that B1, B2, and B12 levels were inversely correlated with FEV₁ and FEV₁/FVC values. There are different results concerning the relation between the antioxidant levels and pulmonary functions in the literature. Rahman et al. [13] found that neither the plasma antioxidant capacity nor protein thiol levels correlated significantly with spirometric data in healthy non-smokers, smokers, or patients with COPD.

Schunemann et al. [19] also found no significant correlation between serum antioxidant capacity measured as TEAC and FEV₁ in a general population, whereas they found an inverse correlation between the levels of lipid peroxidation and lung function. On the contrary, one study showed that total blood glutathione had a significant negative correlation with the severity of airways obstruction (FEV₁% predicted). In the same study, they observed significant differences between the severity of COPD, assessed in terms of reduction in FEV₁% predicted and measures of oxidative stress and antioxidant status [5]. Richards et al. [20] have shown that increased generation of oxidants by peripheral blood leukocytes as measured by luminol-enhanced chemiluminescence was associated with impairment of spirometric parameters. Linden et al. [21] also reported an inverse relationship between bronchoalveolar lavage fluid levels of glutathione peroxidase and airway obstruction measured as FEV₁ in COPD patients. Ochs-Balcom et al. investigated the association between antioxidant nutrients and markers of oxidative stress

with pulmonary function in persons with chronic airflow limitation. They showed that serum beta-cryptoxanthin, lutein/zeaxanthin, and retinol, and dietary beta-carotene, beta-cryptoxanthin, lutein/zeaxanthin, vitamin C, and lycopene were positively associated with FEV₁% and with FVC%. Erythrocytic glutathione was negatively associated with FEV₁%, while plasma thiobarbituric acid-reactive substances (TBARS) were negatively associated with FVC%. They also hypothesized that an imbalance in antioxidant/oxidant status is associated with chronic airflow limitation, and that dietary habits and/or oxidative stress play contributing roles [22].

There has been considerable interest in the association between dietary intake of antioxidants and measurements of systemic oxidative stress and lung function/symptoms in the general population and in smokers [23]. The hypothesis is that a diet rich in antioxidants may help to protect the body against airway inflammation and lung damage caused by cigarette smoke and inhaled environmental pollutants. Thus, individual susceptibility to chronic airways disease may be determined by the ability of the body to deal with inhaled oxidants.

This study confirms the presence of decreased levels of B group vitamins in patients with COPD. The decrease in B vitamin levels correlated with spirometric measurements of airway obstruction in patients with COPD. It may be suggested that B group vitamin supplementation may have a beneficial effect on oxidant/antioxidant imbalance in such patients. Future work is needed to explore the possible relationship between the intake of B group vitamins, systemic and local pulmonary assessment of antioxidant capacity/antioxidant molecules, and lung function.

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