

Article

# Assessment of Vitamin B12 in Patients with Coronary Tortuosity

Vitamin B12 and Coronary Tortuosity

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**Abstract: Background:** Vitamin B 12 deficiency has been implicated in endothelial dysfunction and cardiovascular disease via hyperhomocysteinemia. Coronary tortuosity (CorT) is a common coronary angiography finding. The etiology, clinical implication and long term prognosis are still not well clarified. This study was conducted with the aim to evaluate the relationship between CorT and vitamin B12. **Subjects and Method:** The medical records of consecutive patients, who underwent coronary angiography, were retrospectively reviewed. The study group consisted of 1624 patients. Taking into consideration the inclusion criteria, 212 patients with CorT and 210 patients with normal coronary angiographies (control group) were included in the study. Vitamin B12, other biochemical parameters, clinical and echocardiographic parameters, and CorT score were evaluated in all patients. CorT is defined as fixed 3 bends during both systole and diastole, with each bend  $\geq 45^\circ$ . **Results:** Patients with CorT had higher prevalence of older, female gender, hypertension, current smoking. Vitamin B12 was significantly decreased in patient with CorT ( $134.7 \pm 47.8$  vs  $239.6 \pm 53.8$   $p < 0.001$ ). On multivariate analysis age, female gender, hypertension, diabetes mellitus and vitamin B12 were independent predictors for CorT (OR 1.56; 95% CI: 1.247–1.962;  $p < 0.001$ , OR 1.628; 95% CI: 1.376-2.048;  $p < 0.001$ , OR 1.865; 95% CI: 1.387-2.695;  $p < 0.001$ , OR 1.362; 95% CI: 1.184-1.726;  $p < 0.001$ , OR 1.862; 95% CI: 1.486-2.674;  $p < 0.001$ , respectively). **Conclusion:** In our study, we have founded a significant relationship between vitamin B12 deficiency and CorT.

**Keywords:** vitamin B12; coronary tortuosity; coronary angiography

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## INTRODUCTION

Vitamin B12 is required in the metabolism of homocysteine (Hcy), which occurs through remethylation to methionine or trans-sulfuration to cysteine. Methylation of Hcy, catalyzed by methionine synthetase produces methionine [1]. Hcy-mediated increased lipid peroxidation and generation of free radicals results in inflammation and acute endothelial dysfunction, which accelerates atherosclerotic process predisposing to cardiovascular disease. Coronary artery disease is associated with higher levels of Hcy [2]. Which plays a permissive role in endothelial damage. Low vitamin B12 concentration and hyperhomocysteinemia are common in general [3].

Many studies have been undertaken to examine the relation between plasma Hcy and coronary heart disease [4,5]. The general outcome supports the hypothesis that an elevated plasma Hcy concentration leads to an increased risk of cardiovascular disease.

Coronary tortuosity (CorT) is a common coronary angiography finding. CorT is defined as a fixed 3 bends during both systole and diastole, in at least a single epicardial artery, with each bend  $45^\circ$

change in the vessel direction [6]. The etiology, clinical implication and long term prognosis are still not well clarified. Clinical studies have demonstrated that CorT might be related to aging, hypertension, atherosclerosis and diabetes mellitus [7,8]. The CorT is associated with reversible myocardial perfusion defects and chronic stable angina [9,10]. Zegers et al. [11] demonstrated three cases of patients with the CorT and hypothesized that the CorT may lead to ischemia.

The relationship between vitamin B12 and CorT has not been studied. In the light of these knowledge, we assessed the relationship between the CorT and vitamin B12.

## METHODS

The study group consisted of 1624 consecutive patients, who underwent coronary angiography due to positive treadmill test. A retrospective evaluation of consecutive patients undergoing coronary angiography was performed. Stable angina was defined as discomfort in the chest, back, jaw, shoulder or arms, typically elicited by exertion or emotional stress, and relieved by rest or nitroglycerin. All patients enrolled in the study underwent coronary angiography as a result of chest pain and objective signs of ischaemia during treadmill exercises. Taking into consideration the inclusion criteria, 212 patients with CorT and 210 patients with normal coronary angiographies (control group) were included in the study. We excluded patients with each degree of coronary lesion, previous myocardial infarction.

The data of patients were retrospectively analyzed for the demographic features, echocardiographic parameters, biochemical parameters, vitamin B12 level and CorT.

Vitamin B12 was measured by Ion Capture Microparticle enzyme immunoassay with commercial kits supplied by Abbott Laboratory. Intra assay and inter assay precision was <10% for above parameters. B12 deficiency was defined as <200pg/ml. The study was approved by the local ethics committee.

The patient's data including age, gender, history of hypertension (HT), diabetes mellitus (DM), and echocardiographic variables such as ejection fraction, biochemical and hematologic parameters were received medical records.

### Echocardiographic Examination

All patients underwent transthoracic echocardiography using Vivid S5 (GE healthcare) echocardiography device and Mass S5 probe (2-4 MHz). Standard two-dimensional and color flow Doppler views were acquired according to the guidelines of American Society of Echocardiography and European Society of Echocardiography [12,13]. The ejection fraction was measured according to the Simpson's method.

### Coronary Angiography

All patients underwent elective coronary angiography according to the Judkins technique. Angiograms were reviewed by at least 2 nonblinded reviewing cardiologists. The left anterior descending coronary artery (LAD), the left circumflex coronary artery (LCX) and the right coronary artery (RCA) were observed by various angulations. CorT was evaluated on special angulations, LAD was assessed in right anterior oblique with cranial angulations and LCX in left anterior oblique with caudal angulations, while RCA in right anterior oblique. CorT was identified by  $\geq 3$  bends (defined as  $\geq 45^\circ$  change in vessel direction) along main trunk of at least one artery, present both in systole and in diastole (6).

Patients with DM were identified on admission as those with documented DM using either oral hypoglycemic agents or insulin treatment. Hypercholesterolemia was defined as total cholesterol at least 200 mg/dL or using antihyperlipidemic therapy on admission. Hypertension was defined as blood pressure above 140/90 mmHg or using antihypertensive therapy on admission.

## Statistical Analysis

Statistical analysis was performed using the SPSS (version 20.0, SPSS Inc., Chicago, Illinois) software package. Continuous variables were expressed as mean  $\pm$  standard deviation (mean  $\pm$  SD) and categorical variables were expressed as percentage (%). The Kolmogorov-Smirnov test was used to evaluate the distribution of variables. Student's t-test was used to evaluate continuous variables showing normal distribution, and Mann-Whitney U-test was used to evaluate variables that did not show normal distribution. A p value  $< 0.05$  was considered statistically significant. To identify predictors of CorT, the following variables were initially assessed in a univariate model: age, hypertension, female gender, diabetes, hyperlipidemia, vitamin B12 deficiency. Significant variables in univariate analysis were then entered into a multivariate logistic-regression analysis using backward stepwise selection.

## RESULTS

Prevalance of CorT was found 13% in this study group. Coronary tortuosity and normal coronary group were composed of 212 and 210 patients respectively. The demographic characteristics of both group are summarized in **Table 1**.

There was no significant difference between both groups regarding hyperlipidemia and body mass index (41.5% vs. 40%  $p=0.476$ ,  $26.8 \pm 4.3$  vs.  $24.6 \pm 5.1$   $p=0.549$ ; respectively)(**Table1**)

However, there was a significant difference between both groups regarding hypertension with more hypertensive patients among the CorT group (66.5% vs. 41.9%,  $p<0.001$ )(**Table1**). There were also significant difference between both groups regarding age, female gender, diabetes mellitus ( $68.4 \pm 12.6$  vs.  $56.8 \pm 8.9$   $p<0.001$ , 71.6% vs. 40%  $p<0.001$ , 45.8% vs. 20%  $p<0.001$ ; respectively) (**Table1**).

**Table 1.** General characteristics of patients

Patient Characteristics	Coronary Tortuosity		p
	+	-	
	(212)	(210)	
Age (years)	$68.4 \pm 12.6$ (75)	$56.7 \pm 8.9$ (62)	<b>&lt;0.001</b>
Female gender, n(%)	152 (71.6)	84 (40)	<b>&lt;0.001</b>
Hypertension, n(%)	141 (66.5)	88 (41.9)	<b>&lt;0.001</b>
Diabetes Mellitus, n(%)	97 (45.8)	42 (20)	<b>&lt;0.001</b>
Current smoking, n(%)	132 (62.2)	66 (31.4)	<b>&lt;0.001</b>
Hyperlipidemia, n(%)	88 (41.5)	84 (40)	0.476
Body mass index (kg/m <sup>2</sup> )	$26.8 \pm 4.3$	$24.65.1$	0.549
Statin therapy, n(%)	84 (39.6)	79 (37.6)	0.354

There was no significant difference between both groups regarding urea, creatinine, low density lipoprotein, high density lipoprotein, total cholesterol, triglyceride, uric acid, and folic acid (24.4±7.3 vs. 26.7±6.2 p=0.827, 0.89±0.11 vs. 0.87±0.14 p=0.839, 104±19 vs. 101±24 p=0.864, 41±12 vs. 43±13 p=0.738, 162±24 vs. 157±27 p=0.386, 133±18 vs. 128±19 p=0.842, 5.4±2.1 vs. 5.6±1.9 p=0.751, 9.8±3.3 vs. 10.5±3.9 p=0.763; respectively).

The median serum vitamin B12 level of the CorT group were significantly lower than that of the control group (134.7 ± 47.8 vs. 239.6 ± 53.8, p< 0.001)(Table2).

**Table 2.** Laboratory, and Echocardiographic Parameters

Laboratory and Echocardiographic Variables	Coronary Tortuosity		P
	+	-	
	(212)	(210)	
White Blood Cell (10 <sup>3</sup> / μL)	6.72 ± 1.81 (7.42)	6.93 ± 1.92 (7.51)	0.582
Haemoglobin (g/dl)	13.1 ± 2.3 (14.2)	13.8 ± 1.9 (14.4)	0.895
C Reactive Protein (mg/dl)	0.41 ± 0.16 (0.47)	0.38 ± 0.13 (0.45)	0.627
Total Cholesterol (mg/dl)	162 ± 24 (173)	157 ± 27 (169)	0.386
High Density Lipoprotein (mg/dl)	41 ± 12 (46)	43 ± 13 (48)	0.738
Low Density Lipoprotein (mg/dl)	104 ± 19 (118)	101 ± 24 (116)	0.864
Triglyceride (mg/dl)	133 ± 18 (141)	128 ± 19 (138)	0.842
Urea (mg/dl)	28.4 ± 7.3 (30.4)	26.7 ± 6.2 (29.2)	0.827
Creatinine (mg/dl)	0.89 ± 0.11 (0.92)	0.87 ± 0.14 (0.95)	0.839
Uric acid (mg/dl)	5.4±2.1 (6.1)	5.6±1.9 (6.3)	0.751
Ejection Fraction (%)	62.4 ± 5.2 (64.3)	63 ± 6.1 (65.6)	0.783
Vitamin B12 (pg/ml)	134.7 ± 47.8 (142)	239.6 ± 53.8 (267)	<0.001
Folic acid (ng/ml)	9.8 ± 3.3 (11.8)	10.5 ± 3.9 (12.4)	0.763

The results of univariate analyses are presented in Table 3. On univariate analysis, age, HT, DM, female gender, hprelipidemia, creatinine, smoking, and vitamin B12 were associated with CorT (Table3). On multivariate analysis age, female gender, hypertension, diabetes mellitus and vitamin B12 were independent predictors for CorT (OR 1.56; 95% CI: 1.247–1.962; p < 0.001, OR 1.628; 95% CI: 1.376–2.048; p<0.001, OR 1.865; 95% CI: 1.387–2.695; p<0.001, OR 1.362; 95% CI: 1.184–1.726; p<0.001, OR 1.862; 95% CI: 1.486–2.674; p<0.001, respectively)(Table4).

**Table 3.** Univariate Analysis of Predictors for Coronary Tortuosity

Predictor Variables	OR (95% C.I.)	p
Age (years)	2.138 (1.534 – 3.082)	<0.001
Diabetes mellitus, n(%)	1.857 (1.357 – 2.495)	<0.001
Hypertension, n(%)	2.285 (1.937–2.982)	<0.001
Female gender, n(%)	2.254 (1.962–3.428)	<0.001
Hyperlipidemia, n(%)	1.256 (1.054 – 1.425)	0.001
Creatinine (mg/dl)	1.332 (1.037 – 1.568)	0.001
Current smoking, n(%)	1.426 (1.258–1.692)	0.001
Vitamin B12 (pg/ml)	1.851 (1.375–2.247)	<0.001

**Table 4.** Multivariate Analysis of Predictors for Coronary Tortuosity

Predictor Variables	OR (95% C.I.)	p
Age (years)	1.56 (1.247-1.962)	<0.001
Female gender, n(%)	1.628 (1.376-2.048)	<0.001
Hypertension, (n%)	1.865 (1.387 – 2.695)	<0.001
Diabetes mellitus, (n%)	1.362 (1.184 – 1.726)	<0.001
Vitamin B12 (pg/ml)	1.862 (1.486-2.674)	<0.001

## DISCUSSION

The present study revealed that age, HT, DM, female gender, and vitamin B12 deficiency were associated with CorT. Our results are consisted with previous studies [14-16]. Li et al. [14] found that coronary tortuosity was positively correlated with hypertension. They assumed that the arteries may become tortuous due to reduced axial strain and hypertensive pressure in an elastic cylindrical arterial model. Additionally, some authors claim that CorT is a common finding seen with aging and hypertension due to elongation and dilatation of the arteries associated with left ventricular hypertrophy [15,16].

CorT is a common coronary angiographic finding. There are several possible mechanisms accuse of development CorT. Some authors claimed that degeneration of elastin layer of the vessel may be the cause of coronary tortuosity [17]. CorT might induce subclinical atherosclerosis in the absence of significant obstructive lesions [18]. Severe tortuosity in coronary arteries simplifies atherosclerosis [19]. Hereby, atherosclerosis is more common in patients with coronary artery tortuosity as greater curvature has more areas of low shear wall stress.

Impaired arterial endothelial function and increased carotid intima-media thickness (IMT) were associated with vitamin B12 deficiency. Su et al. [20,21] have found lower vitamin B12, higher Hcy, lower brachial artery resistance in Chinese vegetarians. Kwok et al. [22] reported that strong association between vascular dysfunction and higher plasma Hcy, lower vitamin B12 in Chinese non-smoking vegetarians. Carotid IMT, and brachial flow mediated dilatation were significantly worse in patients with vitamin B12 deficiency than that patients with normal vitamin B12 level [23,24]. Increased cardiovascular risk factors among German vegans and found that vitamin B12 was the only predictor of Hcy concentration [25,26]. Karabudak et al. [27] founden that similar correlation between vitamin B12 and serum Hcy concentration among Turkish women. Vitamin B12 level was the most significant predictor of Hcy among vegans and vegetarians [28].

Studies have illustrated several mechanism for the role of hyperhomocysteinemia in atherogenesis, including auto-oxidation of Hcy, which leads to synthesis of compounds related to the initiation of atherogenic process, such as superoxide, hydrogen peroxide, superoxide anion. These compounds have been implicated in atherogenesis through oxidation of low-density lipoprotein, supression of nitric oxide synthesis, arterial stiffness, endothelial inflammation and foam cell formation [29,30]. Chronic hyperhomocysteinemia reduced activity of superoxide dismutase, which is important in the dismutation of superoxide radicals [30]. Mahalle et al. [31] reported a negative association between vitamin B12 and a positive association between Hcy and triglycerids, very-low-density lipoprotein, and C-reactive protein.

In this light of knowledge, we aim to evaluate relation between CorT and vitamin B12. It's well known that there is a strong association between vitamin B12 deficiency and increased oxidative stress [29-31]. And also, CorT is associated with increased oxidative stress process such as aging, HT, DM, and subclinic atherosclerosis . The present study was the first to evaluate

relationship between CorT and vitamin B12. In our study there was a significant relation between vitamin B12 deficiency and CorT. Further prospective and randomise studies with a larger number of patients are needed on this topic.

### Limitations of the study

Our study has some limitations. First, retrospective study design. Second, small sample size of the present study. Third, the measure of vitamin B12 level was at a single point time. Fourth, the lack of homocysteine levels.

### CONCLUSIONS

To the best of our knowledge, this study is the first to evaluate the relation between CorT and vitamin B12. In our study there was a strong relation between CorT and vitamin B12. Further prospective and randomise studies with a larger number of patients are required due to the relatively small number of patients in the present study.

**Conflict of interest:** None

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