

■ Research Article

## Repolarization changes associated with serum vitamin B12 levels: evaluation with frontal QRS-T angle

### *Serum vitamin B12 düzeyleri ile ilişkili repolarizasyon değişiklikleri: frontal QRS-T açısı ile değerlendirme*

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### Abstract

**Aim:** Vitamin B12 deficiency may influence cardiac autonomic function and ventricular repolarization. The frontal QRS-T angle is a novel and useful electrocardiographic parameter that reflects the relationship between ventricular depolarization and repolarization. The aim of this study was to evaluate the association between serum vitamin B12 levels and the frontal QRS-T angle in asymptomatic individuals.

**Material and Methods:** The current retrospective study comprised 92 adults who underwent a standard 12-lead ECG and serum vitamin B12 testing on the same day. Participants were divided into two groups according to their vitamin B12 levels (Group 1 was defined as abnormal having a level of less than 300 pg/mL, while Group 2 was defined as normal having a level of 300 pg/mL or more). The patient's clinical characteristics, laboratory findings and ECG parameters were recorded. The frontal QRS-T angle was calculated automatically using the QRS and T axes provided by the ECG device.

**Results:** The average age of participants was 46.1 ( $\pm 12.2$ ) years, and 51.9% of participants were female ( $p < 0.001$ ). Individuals with low vitamin B12 concentrations exhibited a wider frontal QRS-T angle. It was determined that vitamin B12 level is an independent predictive factor for the frontal QRS-T angle.

**Conclusion:** The frontal QRS-T angle may serve as a useful parameter for evaluating electrical changes and susceptibility to ventricular arrhythmias in individuals with vitamin B12 deficiency.

**Keywords:** electrocardiography, frontal QRS-T angle, repolarization, vitamin B12 level

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## Öz

**Amaç:** Vitamin B12 eksikliği, kardiyak otonomik fonksiyonu ve ventriküler repolarizasyonu etkileyebilir. Frontal QRS-T açısı, ventriküler depolarizasyon ve repolarizasyon arasındaki ilişkiyi yansıtan yeni ve kullanışlı bir elektrokardiyografik parametredir. Bu çalışmanın amacı, asemptomatik bireylerde serum vitamin B12 düzeyleri ile frontal QRS-T açısı arasındaki ilişkiyi değerlendirmektir.

**Gereç ve Yöntemler:** Mevcut retrospektif çalışma, aynı gün standart 12 derivasyonlu EKGsi çekilen ve serum B12 vitamini düzeyi bakılan 92 yetişkin kapsamaktadır. Katılımcılar, B12 vitamini düzeylerine göre iki gruba ayrıldı. (Grup 1, 300 pg/mL'den düşük düzeyde olan Grup 2, 300 pg/mL veya daha yüksek düzeyde olarak tanımlanmıştır). Hastaların klinik özellikleri, laboratuvar bulguları ve EKG parametreleri kaydedildi. Frontal QRS-T açısı, EKG cihazı tarafından sağlanan QRS ve T eksenleri kullanılarak otomatik olarak hesaplandı.

**Bulgular:** Katılımcıların ortalama yaşı 46,1 ( $\pm 12,2$ ) idi ve katılımcıların %51,9'u kadındı ( $p < 0,001$ ). Düşük serum vitamin B12 konsantrasyonuna sahip bireylerin daha geniş frontal QRS-T açısına sahip olduğu görüldü. Serum Vitamin B12 seviyesinin frontal QRS-T açısı için bağımsız bir prediktif faktör olduğu gösterildi.

**Sonuç:** Frontal QRS-T açısı, vitamin B12 eksikliği olan bireylerde subklinik elektriksel değişiklikleri değerlendirmek için yararlı bir parametre olarak kullanılabilir.

**Anahtar kelimeler:** elektrokardiyografi, frontal QRS-t açısı, repolarizasyon, vitamin B12

## Introduction

Vitamin B12 (cobalamin), is a water-soluble molecule and a vital component of the human body. Vitamin B12 and its coenzyme methyl-malonyl-Co-A mutase plays a pivotal role in DNA synthesis, myelin synthesis, lipid and carbohydrate metabolism. It is essential for the proper functioning of the hematological and neurological systems and the maintenance of optimal neurological function [1,2].

As is well documented in the literature, vitamin B12 deficiency is associated with macrocytic (or megaloblastic) anaemia, pancytopenia, neurological consequences, including paresthesias, peripheral neuropathy, and demyelination which can frequently be reversed by early diagnosis and appropriate treatment [2]. Vitamin B12 has been demonstrated to have significant effects on the sympathetic and parasympathetic systems. Consequently, symptoms of autonomic dysfunction, including orthostatic hypotension and decreased heart rate variability, may manifest in cases of B12 deficiency. Notably, these autonomic dysfunction manifestations can sometimes occur prior to the manifestation of neurological or haematological abnormalities. Moreover, the impact of B12 deficiency on the sympathetic system is more pronounced than its effect on the parasympathetic system [1-3].

Ventricular repolarization (VR) parameters, including the QT interval, and the corrected QT interval value (QTc), QT dispersion (QTd), which can be measured using surface electrocardiography (ECG), have frequently been used in studies which aim to predict the risk of potentially lethal ventricular arrhythmias [4].

The frontal QRS-T angle [f(QRS-T)] which is defined as the angle between ventricular depolarization (QRS wave) and repolarization (T wave), has recently been accepted as an indicator of cardiac electrical activity. The f(QRS-T) angle can be readily obtained by calculating the absolute difference between the QRS and T axes from a 12-lead digital surface ECG. The frontal method and the spatial method are two established methods that are used to calculate the f(QRS-T) angle. The frontal method is a straightforward calculation method that employs the automatic ECG report generated by the ECG device. It has been demonstrated that this method exhibits a robust correlation with the more intricate and sophisticated spatial QRS-T angle [5]. In conditions conducive to optimal functioning, the angle between the QRS complex and the T wave should be relatively narrow, indicating that depolarization and repolarization occur simultaneously. This observation lends further credence to the hypothesis that cardiac electrical conduction and repolarization abnormalities are associated with an increase in the f(QRS-T) angle.

As demonstrated in preceding studies, an augmentation in the f(QRS-T) angle has been identified as a promising independent prognostic indicator for the emergence of diverse cardiovascular pathologies, notably malignant arrhythmia and sudden cardiac death [5-8]. Despite the established relationship between arrhythmia risk and vitamin B12 deficiency, as evidenced by prior studies, the potential association between vitamin B12 deficiency and the f(QRS-T) angle remains to be delineated [9]. The objective of the present study is to determine the relationship between vitamin B12 deficiency and the f(QRS-T) angle.



## Material and Methods

The present retrospective, single-centre, cross-sectional study was designed to evaluate the association between the f(QRS-T) angle and serum vitamin B12 levels. A total of 92 consecutive patients who presented to the cardiology outpatient clinic for routine evaluation between December 2024 and March 2025 were included in the study. Participants were eligible for inclusion in the study if they were between 18 and 65 years of age and if both a standard 12-lead ECG and a serum vitamin B12 measurement had been performed on the same day. It is noteworthy that none of the patients exhibited symptoms and none were receiving vitamin B12 supplementation at the time of assessment. Individuals with a history of cerebrovascular disease, active infection, chronic kidney disease, myocardial infarction, moderate to severe valvular pathology, heart failure, electrolyte imbalance, malignancy, or any disorder known to affect cardiac conduction or repolarization were excluded from the study. The ECG artefacts, bundle branch blocks, atrial fibrillation and pacemaker rhythm constituted additional exclusion criteria.

The clinical and demographic characteristics of the participants, including age, sex, and comorbidities such as hypercholesterolaemia, diabetes mellitus, and hypertension, were meticulously documented. Patients who had smoked within the preceding 12 months were classified as current smokers. The body mass index (BMI) was calculated by dividing the subject's weight in kilograms by their height in metres squared.

### Laboratory Data

A comprehensive laboratory assessment was performed for each participant. The complete blood count parameters, liver and renal function tests, serum electrolytes, thyroid function tests, and C-reactive protein (CRP) were all included in the study. The measurement of vitamin B12 concentrations was conducted using venous samples, which were analysed with the Architect Vitamin B12 assay kit (Abbott Laboratories, Chicago, USA). In accordance with contemporary literature demonstrating haematological and neurological symptoms between 200–300 pg/mL and proposing this range as a borderline threshold for functional deficiency, patients were categorised into two groups based on serum vitamin B12 level: Group 1 was defined as abnormal having a level of less than 300 pg/mL, while Group 2 was defined as normal having a level of 300 pg/mL or more [10,11].

### Electrocardiography

A standard 12-lead ECG was recorded in the supine position after adequate rest, using the Beneheart R12 (Mindray) device with settings of 10 mm/mV and 25 mm/s. The ECGs were subsequently scanned and transferred to the workstations of two

independent cardiologists who were blinded to the patient data. This approach was adopted to minimise potential measurement bias, particularly for parameters such as QT, QTc, QRS, P-wave dispersion (Pd), Tp e, Tp e/QTc, and the frontal QRS-T angle. Electrocardiographic images were magnified fourfold using Adobe Photoshop to enhance measurement precision.

The Tp e interval, representing transmural dispersion of repolarization, was measured from the peak to the end of the T wave across 12 leads [5]. The QT interval was defined as the time from the onset of the QRS complex to the termination of the T wave, and QTc was calculated using Bazett's formula: QTc = QT /  $\sqrt{R\text{R interval}}$  [6]. Electrocardiograms (ECGs) displaying prominent U waves were excluded from the study to avoid interpretative interference.

The f(QRS-T) angle was obtained by subtracting the T wave axis from the QRS axis in the frontal plane. In instances where the resulting value exceeded 180°, it was subjected to a correction process involving the subtraction of 360°. In order to reduce variability attributable to the observer and increase measurement reliability by means of standardisation, the value automatically obtained by the ECG device as the f(QRS-T) angle was used.

### Echocardiographic Assessment

Transthoracic echocardiography (TTE) was performed by two cardiologists with extensive experience in this field, and both were unaware of the findings of the other. The ultrasound system utilised for the examinations was a Vivid S5 (GE Vingmed Ultrasound AS, Horten, Norway), and the procedures were performed in accordance with the current guidelines [7]. The echocardiographic assessment incorporated conventional M mode and two dimensional imaging, in addition to pulsed wave and colour Doppler evaluations. The left ventricular ejection fraction (LVEF) was calculated using the modified Simpson method.

This study was approved by the Ethics Committee of Medicalpark Hospital, Antalya, Turkey on 22 August 2024 with approval code number 2024/7. The study was conducted in line with the principles of Declaration of Helsinki and Good Clinical Practice, with the aim of respecting the rights and dignity of all parties involved. All participants were provided with detailed information about the purpose and the protocol of the study, after which verbal informed consent was taken from all of them.

### Statistical Analysis

Statistical analysis were performed using JAMOVI, a third-generation, open-source, R-based software. Descriptive statistics were presented as mean  $\pm$  standard deviation (SD) or median and interquartile range (IQR), as appropriate. In the vitamin B12 subgroup, mean comparisons were conducted using the Student's t-test. Categorical variables

were compared using the chi-square test. For non-parametric variables, medians were analyzed with the Wilcoxon test. The Kolmogorov-Smirnov or Shapiro-Wilk tests were used to assess whether the variables followed a normal distribution. To evaluate the relationship between the f(QRS-T) angle and serum vitamin B12 levels, Pearson or Spearman correlation coefficients were applied depending on distribution

characteristics. To identify the best predictors of the f(QRS-T) angle, a multivariate linear regression analysis using the enter (input) method was performed. A p-value <0.05 was considered statistically significant (Table 1). In the regression analyses, vitamin B12 level emerged as the sole independent predictor of the frontal QRS-T angle ( $\beta = 0.035$ , 95% CI: 0.008–0.062,  $p = 0.013$ ), with no other variables maintaining statistical significance after adjustment (Table 2).

**Table 1.** Baseline demographic and clinical features of the study.

Variable	Vitamin B12 <300 pg/mL (n=55)	Vitamin B12 ≥300 pg/mL (n=37)	Total (n=92)	p-value
Age, years	46.0 ± 12.3	46.3 ± 12.2	46.1 ± 12.1	0.71
Female sex, n (%)	28 (50.9)	20 (54.1)	48 (52.2)	0.76
BMI, kg/m <sup>2</sup>	26.2 ± 5.0	26.2 ± 4.2	26.2 ± 4.7	0.93
Hypertension, n (%)	16 (29.1)	12 (32.4)	28 (30.4)	0.72
Diabetes mellitus, n (%)	16 (29.1)	7 (18.9)	23 (25.0)	0.27
Dyslipidemia, n (%)	5 (9.1)	7 (18.9)	12 (13.0)	0.18
Current smoking, n (%)	36 (65.5)	29 (78.4)	65 (70.7)	0.19
Coronary artery disease, n (%)	3 (5.5)	2 (5.4)	5 (5.4)	0.99
Glucose, mg/dL	105.9 ± 37.1	100.6 ± 31.8	103.9 ± 35.3	0.34
Creatinine, mg/dL	0.80 ± 0.20	0.79 ± 0.17	0.80 ± 0.20	0.60
Hemoglobin, g/dL	14.2 ± 1.8	14.4 ± 1.5	14.3 ± 1.7	0.62
TSH, mIU/L	1.8 (1.2–2.6)	1.9 (1.1–2.5)	1.9 (1.2–2.5)	0.66
LVEF, %	62.4 ± 2.5	62.6 ± 2.5	62.5 ± 2.5	0.88
LA diameter, mm	35.2 ± 4.3	35.4 ± 4.5	35.3 ± 4.4	0.84
LVDD, mm	46.0 ± 3.3	45.8 ± 3.8	45.9 ± 3.7	0.44
IVS thickness, mm	9.8 ± 1.9	9.6 ± 2.2	9.7 ± 2.0	0.56
Posterior wall thickness, mm	9.7 ± 2.1	9.6 ± 2.3	9.7 ± 2.2	0.74
QT dispersion, ms	39.6 ± 7.2	40.7 ± 6.2	40.0 ± 6.9	0.28
QTc dispersion, ms	43.3 ± 7.7	44.2 ± 6.6	43.8 ± 7.3	0.29
Tp-e interval, ms	74.5 ± 11.3	74.5 ± 10.7	74.5 ± 11.1	0.99
Tp-e/QTc ratio	0.19 (0.16–0.20)	0.19 (0.17–0.20)	0.19 (0.16–0.20)	0.95
QRS duration, ms	88.1 ± 9.2	87.5 ± 8.8	87.9 ± 9.0	0.69
QTc interval, ms	405.9 ± 19.2	405.7 ± 20.0	405.8 ± 19.5	0.95
Frontal QRS-T angle, °	23.5 (14–46.2)	8 (4–27)	19 (8.5–37)	<0.001

1:Pearson's Chi-squared test, 2:Lineer model ANOVA, 3:Kruskal-Wallis test, SD: Standard Deviation, ALT: Alanine Aminotransferase, AST : Aspartate Aminotransferase, BMI: Body Mass Index, CAD: Coronary Artery Disease, f(QRS-T): Frontal QRS-T angle, HDL: High-Density Lipoprotein Cholesterol, IQR: Interquartile Range, LDL: Low-Density Lipoprotein Cholesterol, Pd: P-wave duration, QRS: QRS duration, QTc: Corrected QT interval QTcd: Corrected QT interval dispersion, TSH: Thyroid Stimulating Hormone, Tp-e: Interval from the peak to the termination of the T-wave, Tp-e/QTc: Ratio of Tp-e to corrected QT interval, \*: Group 1= Vitamin B12 < 300 pg/ml, Group 2= Vitamin B12 > 300 pg/ml

**Table 2.** Univariate and Multivariate Linear Regression Analysis of Predictors for frontal QRS-T angle.

Variables	Univariate				Multivariate			
	B	p-value	Lower	Upper	$\beta^*$	p-value	Lower	Upper
p-value	Lower	Upper						
Age	0.236	0.270	-0.186	0.658	-	-	-	-
BMI	0.473	0.393	-0.623	1.569	-	-	-	-
Hemoglobin	-0.495	0.724	-3.269	2.279	-	-	-	-
Hypertension	4.116	0.573	-10.167	18.399	-	-	-	-
Diabetes	10.331	0.205	-5.770	26.432	-	-	-	-
LV EF	-0.190	0.960	-0.752	0.715	-	-	-	-
B12	0.038	0.031	0.004	0.073	0.035	0.013	0.008	0.062
CAD	0.055	0.750	-0.102	0.173	-	-	-	-

B: unstandardized regression coefficient, BMI: Body mass index, LV EF: Left ventricular ejection fraction, TSH: Thyroid-stimulating hormone, CAD: Coronary artery disease,  $\beta$ : standardized regression coefficient, \*: F:2.347, R2:0.230, p<0.001"

## Results

Table 1 provides a summary of the baseline demographic, clinical, electrocardiographic (ECG), and echocardiographic characteristics of the study population for various vitamin B12 levels. The total number of participants included in the study was 92, of whom 51.9 % were female (p<0.001). The mean age of the study group was 46.1 years ( $\pm 12.1$ ). No significant differences were observed among the participants among the various vitamin B12 level groups (p>0.05 for all). The ECG parameters including p wave duration, QT interval, QTc interval dispersion, QRS duration, interval from peak of the T wave to its termination, and the ratio of Tp-e to QTc, remained consistent across all subgroups (p>0.05 for all). However, the f(QRS-T) angle exhibited a propensity to rise in cases of decreased vitamin B12 levels. (p<0.05). The study found that vitamin B12 levels had no significant impact on the majority of echocardiographic parameters.

## Discussion

The present study demonstrates a significant association between lower serum vitamin B12 concentrations and an increased f(QRS-T) angle, suggesting that vitamin B12 deficiency may influence the spatial heterogeneity of ventricular depolarization and repolarization. To the best of our knowledge, this is the first study to investigate the relationship between vitamin B12 levels and the f(QRS-T) angle. This parameter has gained growing recognition as a sensitive marker of arrhythmic risk and adverse cardiovascular outcomes [4,6].

Vitamin B12 plays a pivotal role in a number of vital bodily functions, including DNA synthesis, myelin formation,

erythropoiesis, and mitochondrial function. As is well established, the absence of this vitamin B12 has been shown to induce a number of serious health complications, including macrocytic anaemia, pancytopenia, demyelination, autonomic dysfunction and peripheral neuropathy. As demonstrated in previous studies, vitamin B12 deficiency has been shown to affect both the sympathetic and parasympathetic systems, with a more pronounced impairment of the sympathetic system. Autonomic dysregulation has been linked to abnormalities in ventricular repolarization, potentially serving as a mechanistic bridge between B12 deficiency and increased arrhythmic vulnerability [3,9].

The f(QRS-T) angle reflects the alignment between depolarization and repolarization vectors. A wider angle indicates increased electrical heterogeneity across the myocardium and has been associated with a higher risk of malignant ventricular arrhythmias and sudden cardiac death. While traditional ECG parameters such as QT interval, QTc, and Tp-e have been used to assess repolarization, the f(QRS-T) angle is emerging as a robust predictor of global electrical instability. Our findings suggest that vitamin B12 deficiency may contribute to these repolarization abnormalities.

Prior research has primarily focused on the effects of vitamin B12 deficiency on QT prolongation or autonomic dysfunction. Yilmaz et al showed that low vitamin B12 levels in healthy individuals may cause repolarization disorders that increase arrhythmogenic susceptibility [9]. Although the literature includes evidence linking B12 deficiency to arrhythmic predisposition through autonomic imbalance, no previous

investigation has explored the f(QRS-T) angle in this context, making our study an important contribution to the field.

Although universal screening is not recommended, testing should be considered in patients with one or more risk factors, such as gastric or small intestine resections, inflammatory bowel disease, use of metformin for more than four months, use of proton pump inhibitors or histamine H2 blockers for more than 12 months, vegans or strict vegetarians, and adults older than 75 years. Initial laboratory assessment should include a complete blood count and serum vitamin B12 level. A serum B12 above 300 pg/mL is interpreted as normal. Patients with B12 levels between 200 and 300 pg/mL are considered borderline, and measurement of serum methylmalonic acid may be helpful in diagnosis [10,11].

Vitamin B12 deficiency can disrupt cardiac autonomic modulation through several pathways, including impaired myelin synthesis and autonomic imbalance. Elevated homocysteine levels can contribute to endothelial dysfunction and adverse effects on myocardial conduction. Mitochondrial dysfunction related to impaired methyl-malonyl-CoA metabolism may also affect the electrophysiological stability of ventricular myocytes [11,12].

In multivariate regression analysis, serum vitamin B12 level emerged as the sole independent predictor of the f(QRS-T) angle. This finding should be cautiously interpreted as the relationship was small and of limited clinical significance. Although the positive standardized beta coefficient indicates a direct relationship, the magnitude of the effect was small, suggesting limited clinical impact at the individual level.

In daily cardiology practice, many patients receive long-term antiplatelet and anticoagulant therapies, often in combination with chronic proton pump inhibitor (PPI) therapy, with the aim of reducing the risk of gastrointestinal bleeding. Large population studies have demonstrated that PPI use over two years increases the likelihood of deficiency. Clinical guidelines also recommend reassessing B12 status for those on chronic PPI or H2-blocker therapy. In light of the findings of the present study, it may be considered prudent to implement a routine monitoring procedure for vitamin B12 levels in cardiology patients requiring long-term PPI therapy. Early identification and correction of B12 deficiency may help mitigate ventricular repolarization abnormalities and reduce arrhythmic susceptibility. It can thus be concluded that the routine assessment of vitamin B12 levels could represent a simple, non-invasive, and cost-effective strategy within the context of comprehensive cardiovascular care.

### Limitations of the study

The retrospective, cross-sectional design of the study

precludes the drawing of causal inferences. The study did not include the measurement of functional biomarkers such as methylmalonic acid and homocysteine, which may have provided more clarity regarding metabolic deficiency in borderline cases. The study's limitations include the lack of 24-hour electrocardiographic monitoring. The f(QRS-T) angle provides info on ventricular repolarization heterogeneity, but not on arrhythmic burden or heart rate variability. Long-term rhythm monitoring could enable a more comprehensive evaluation of subclinical arrhythmias and autonomic function. Moreover, the study population consisted of relatively healthy outpatients, which limits the generalisability of the results. This study has several strengths, including careful ECG measurements, appropriate exclusion of confounding cardiac conditions, and adherence to contemporary diagnostic standards for vitamin B12 deficiency. Despite these limitations, the present findings highlight an important and potentially modifiable determinant of ventricular electrical stability. The conduction of prospective studies with larger populations is indicated to confirm these results and to explore whether vitamin B12 supplementation can reduce the f(QRS-T) angle and improve arrhythmic outcomes.

In conclusion, in this study, vitamin B12 level was identified as the only independent predictor of the frontal QRS-T angle, indicating heightened ventricular electrical heterogeneity. These findings underscore the importance of recognising and correcting vitamin B12 deficiency, particularly in cardiology patients exposed to long-term PPI therapy, and suggest that maintaining adequate vitamin B12 levels may serve as a preventive strategy against arrhythmic risk.

### Declaration of conflicting interests

The authors declare they have no conflicts of interest.

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### Ethics approval

This study was approved by the Ethics Committee of Medcalpark Hospital, Antalya, Turkey on 22 August 2024 with approval code number 2024/7.

### Authors' contribution

FAD: Concept, Design, Analysis and/or Interpretation, Writing of the Article. GB: Design, Materials, Data Collection and/or Processing, Critical Review. AA: Data Collection and/or Processing, Literature Review. MD: Materials, Data Collection and/or Processing, Analysis and/or Interpretation. ÖÖK: Data Collection and/or Processing, Literature Review. SÜ: Supervision, Analysis and/or Interpretation, Critical Review.



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