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The effect of cigarette smoking on serum homocysteine, folic acid, and vitamin B12 concentrations in patients with cardiovascular diseases

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Abstract

Background: Smoking is recognized as a significant risk factor for cardiovascular diseases (CVD), yet its influence on blood levels of homocysteine, folic acid, and vitamin B12 is not well understood. This study aimed to explore how smoking affects these biochemical markers in patients with CVD.

Methods: The study included 88 participants diagnosed with CVD, who were categorized into smokers (n=44) and non-smokers (n=44). Serum concentrations of homocysteine, folic acid, and vitamin B12 were assessed using ELISA. Additionally, blood pressure (both systolic and diastolic) and body mass index (BMI) were recorded.

Results: Smokers showed significantly lower levels of folic acid (22.41 ± 5.95 ng/mL) compared to non-smokers (28.05 ± 4.13 ng/mL, p = 0.000). No significant differences were observed in homocysteine (p = 0.958) or vitamin B12 (p = 0.578) levels between the two groups. A negative correlation was found between folic acid and systolic blood pressure in smokers, while no significant associations were noted among folic acid, vitamin B12, and homocysteine.

Conclusion: In patients with CVD, smoking is linked to significantly lower folic acid levels, which may lead to increased systolic blood pressure. These results underscore the need to monitor folic acid levels in smokers who are at risk for cardiovascular issues.

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Introduction

Cigarette smoking is considered a serious health concern that has adverse effects on human well-being. There is a strong link between smoking and the progression of heart-related diseases (1,2). Smoking contains thousands of toxic compounds that may have detrimental effects on cardiovascular health. Interestingly, a significant number of preventable deaths in developed and prosperous areas of the world are attributed to smoking. While a robust epidemiological link exists between smoking and cardiovascular disease (CVD), the mechanisms for developing of CVD in smokers remain unclear (3). Vascular endothelial damage is considered a fundamental step in the case of atherosclerosis pathogenesis (3). Accumulation of reactive oxygen species (ROS), along with oxidative events is a key sector in vascular endothelial dysfunction following exposure to smoking (4).

Homocysteine is a sulfur-containing metabolite generated from methionine by the loss of a terminal methyl group (5). Comprehensive research in European populations has shown that smoking is associated with elevated plasma homocysteine levels. Moreover, higher homocysteine concentrations have been linked with an increased risk of CVD, possibly through mechanisms such as oxidative stress, inflammation, and endothelial dysfunction (1). On the other hand, smoking is associated with an insufficient intake of vitamins. In this regard, a lack of folate and vitamin B12 is commonly observed in a significant number of hyperhomocysteinemia patients (6). Notably, administration of B vitamins, such as folic acid, can reduce homocysteine levels and prevent the development of CVD (5). In fact, exposure to smoking can lead to diminished intake of folic acid and vitamin B12, which are necessary for the normal function of metabolic enzymes (7).

Given the harmful effects of smoking on various aspects of human health, we aimed to evaluate the impact of cigarette smoking on serum homocysteine, folic acid, and vitamin B12 levels in patients with atherosclerosis. The findings of this study may provide valuable insights into the mechanisms underlying the association between cigarette smoking and CVD risk and may have important implications for developing strategies to prevent and manage CVD in high-risk populations.

Methods

The current study was a case-control study conducted from November to April 2022-23 at the Metabolic Disorder Research Centre in Gorgan, Golestan Province, Iran. The study was approved by the Ethics Committee (No: IR.GOUMS.REC.1401.534) of the Golestan University of Medical Sciences. Eighty-eight patients with cardiovascular complications were included in this study. All participants gave their informed consent. The non-smoker group (n= 44) included patients reporting to be never smokers. The smoker group (n= 44) consistent of patients who consumed 5-15 cigarettes daily. The number of cigarettes was estimated by the patient at intake, confirmed by her partner. Exclusion criteria were chronic hypertension, familial dyslipidemia, chronic heart disease, and the use of folic acid or vitamin supplements. Blood samples were drawn between 8.30 and 9.30 a.m. after fasting overnight for 10 h and a resting period of 20 min and in the smoker's group at least 1 h after the last cigarette was smoked. Five-milliliter blood samples were provided for all subjects after a 12-h overnight fasting. After serum separation, it was used to determine biochemical parameters. Immediately after venesection, the plasma was separated, stored at -70 ºC in plastic tubes, and thawed in 37 ºC water for 5 min before serial analysis. Serum homocysteine (Zell Bio, Germany), vitamin B12, and folic acid (Monobind, USA) levels were determined with an enzyme-linked immunosorbent assay (ELISA).

Weight was measured using a digital weight balance. Heavy clothing was removed, and participants wore light clothing only, with an estimated weight reduction of about 0.5 Kg. Body mass index (BMI) was calculated by dividing weight (Kg) by height squared (m2). Systolic blood pressure (SBP) and diastolic blood pressure (DBP) (mmHg) were

measured a resting period of approximately 5 min. The student t-test was used to compare the differences between the mean values of the parameters in both groups at p <005. Normality was assessed by Kolmogorov-Smirnov test using SPSS software, version 21. For the calculation of the coefficients of correlation, the Spearman rank test was used.

Results

A total of 44 cases and 44 age- and sex-matched controls were analyzed. The mean age of smokers and non-smokers was 62.10 ± 9.97 and 65.46 ± 8.98, respectively (p=0.096). Additionally, 31 patients (70.2%) and 24 controls (54.4%) were male (p=0.3). The smoker group showed a significant reduction in serum folic acid relative to the non-smoker group (p<0.001) (Table 1). Smoker patients had significantly higher SBP and DBP than non-smokers. There was also a decrease in serum vitamin B12 levels in the smoker's group, although not significantly. In addition, a non-significant increase was observed in body mass index and homocysteine levels compared to the control group. To examine the relationship between folic acid and study parameters, we also performed a correlation study. There is a moderate, negative, and significant correlation between folic acid and systolic blood pressure in the smoker group (r=-0.293, p=0.048). We also found no significant association between folic acid and study parameters in both groups (Table 2).

Table 1. Demographic and biochemical characteristics of the study population

Parameter	Mean ± SD		P-Value
	Smokers (n=44)	Non-smokers (n=44)	
Age (Years)	62.10 ± 9.97	65.46 ± 8.98	0.096
BMI (Kg/m^2)	27.47 ± 2.70	27.28 ± 4.80	0.583
DBP (mmHg)	80.13 ± 10.74	70.77 ± 10.74	0.025*
SBP (mmHg)	130.54 ± 11.42	120.72 ± 11.40	0.009*
Homocysteine (nm/ml)	32.33±10.9	32.22±8.61	0.958
Folic acid (ng/ml)	22.41±5.95	28.05±4.13	0.001*
Vitamin B12 (ng/ml)	43.97±8.02	47±6.88	0.578

Data were presented as mean ± standard deviation (SD). Student t-test was applied. *p<0.05 is considered significant. BMI: Body Mass Index; DBP: Diastolic Blood Pressure; SBP: Systolic Blood Pressure. P-value indicates the statistical difference between non-smokers and smokers.

Table 2. Correlations of folic acid with study parameters in smoker and non-smoker individuals

Parameter	Smokers (n=44)		Non-smokers (n=44)	
	r	P-Value	r	P-Value
Age (Years)	0.133	0.214	- 0.16	0.298
BMI (Kg/m2)	0.110	0.467	- 0.018	0.907
DBP (mmHg)	- 0.095	0.530	- 0.149	0.335
SBP (mmHg)	- 0.293*	0.048*	- 0.104	0.502
Homocysteine (nm/ml)	0.089	0.557	0.094	0.542
Vitamin B12 (ng/ml)	- 0.156	0.299	0.260	0.088

Correlation-Spearman tests were applied. *p<0.05 is considered significant. BMI: Body Mass Index; DBP: Diastolic Blood Pressure; SBP: Systolic Blood Pressure.

Discussion

Research on the effects of cigarette smoking on human well-being is vital to demonstrate the dangers of smoking. In this study, we investigate the impact of cigarette smoking on serum homocysteine, folic acid, and vitamin B12 levels in smoker and non-smoker patients with CVD.

Our results indicate a significant reduction in serum folic acid levels in smokers compared with non-smokers. This finding is consistent with previous studies that reported a negative connection between cigarette smoking and serum folic acid levels (8,9). The underlying reason for this association is not well understood; however, it may be due to the fact that harmful chemicals present in cigarette smoke lead to increased oxidative stress, which in turn can lead to the breakdown of folic acid in the body. In addition, smokers often consume less fruit and vegetables, which are the primary sources of these vitamins, which could lower serum levels (7). Based on a study conducted by Vardavas et al., smoking status is associated with lower consumption of fiber, fruit, and vegetables, and smoking status affects serum folic acid regardless of diet (10).

In addition, our findings showed a non-significant reduction in serum vitamin B12 levels in the smokers group. This could be due to the small number of the participants. Our findings also indicate a non-significant change in serum homocysteine levels between smokers and non-smokers. In contrast to our findings, several reports have demonstrated that smoking can increase serum homocysteine levels (11). Based on previous studies, in addition to the sample size, factors such as the number of cigarettes smoked per day, duration of smoking status, age, gender, and consumption of coffee and exercise may influence homocysteine levels in serum (7). On the other hand, patients with CVD often experience high serum homocysteine levels, which may mask any differences in homocysteine levels associated with smoking in our study, which can be considered another possible explanation.

The results from the correlation study indicate a negative significant association between serum folic acid and systolic blood pressure in smoking individuals. This result suggests that lower levels of folic acid may contribute to an increase in systolic blood pressure among smokers. However, we did not observe a significant correlation between folic acid levels and diastolic blood pressure. In order to confirm the correlation between folic acid and hypertension, further studies with a larger sample size are needed. The relationship between folate intake and hypertension has been studied extensively. The reason why low folic acid levels give rise to elevated blood pressure is often related to the augmented deposition of homocysteine in blood vessels, which subsequently leads to endothelial dysfunction. Homocysteine has been linked to arteriolar constriction, increased sodium reabsorption, renal dysfunction, and increased arterial stiffness, all of which can lead to elevated blood pressure (12,13). Furthermore, we did not observe any significant relationship between folic acid and vitamin B12, as well as homocysteine in this study.

Conclusion

The findings of the present study suggest that smoker patients with a low level of folic acid may experience elevated blood pressure. This highlights the potential benefits of folic acid supplementation for heart failure patients. However, we found no significant changes in serum homocysteine and vitamin B12 levels between the two groups. There may be some limitations to sample collection, technical methods, or variations in the amount of cigarette smoke exposure that can affect these results.

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Ethical statement

Ethical approval for this research study was granted by the Golestan University of Medical Sciences Ethics Committee (No. IR.GOUMS.REC.1401.534). All procedures were performed in accordance with the guidelines for studies involving human participants, considering the ethical standards of the institutional and/or national research committee, as well as the 1964 Helsinki Declaration and its later amendments or comparable ethical standards. During data collection, the researchers obtained informed consent from the participants after explaining the purpose and objectives of the study.

Conflicts of interest

The authors declare that they have no conflict of interest.

Author contributions

HRJ and ShH conceptualized the study, provided the project design, and interpreted the data. NB analyzed the data. KhGh contributed to collecting the serum and data. ZH and FF interpreted the data, drafted, and wrote the manuscript. NH collected the data and contributed to writing the manuscript. All authors read and approved the final version of the manuscript.

Data availability statement

The data supporting the findings of this study are available from the corresponding author upon reasonable request.

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