Correlation Between Serum Homocysteine Levels and Carotid Intima-media Thickening in Hemodialysis Patients

Alireza Soleimani,[1] Seyed Hamed Tabatabaei,[1]
Maryam Soleimani,[2] Mehrdad Sinaeinejad,[3] Fereshteh Ghazvini,[4]
Seyed Zia Tabatabaei,[5] Hossein Mofidi,[6] Mehdi Kashani,[7]
Seyyed Taher Seyyed Mahmoudi[8]

[1]Internal Medicine Department, Kashan University of Medical Sciences, Kashan, Iran
[2]Ankara University faculty of pharmacy, Ankara, Turkey
[3]Internal Medicine Department, Qom University of Medical Sciences, Qom, Iran
[4]Yazd University of Medical Sciences, Yazd, Iran
[5]Internal Medicine Department, Sina Hospital, Tehran University of Medical Sciences, Tehran, Iran
[6]Faculty of Industrial Engineering, Science and Research Branch, Islamic Azad University, Tehran, Iran
[7]School of Medicine, Tehran University of Medical Sciences, Tehran, Iran
[8]School of Medicine, Tabriz University of Medical Sciences, Tabriz, Iran

Keywords. carotid intima-media thickness, hemodialysis, atherosclerosis, homocysteine

Original Paper

Introduction. Hyperhomocysteinemia is an important risk factor for cardiovascular disease in ESKD patients. Homocysteine, as an inflammatory factor, and carotid intima-media thickness (CIMT) could predict atherosclerosis in hemodialysis-treated ESKD patients. In this regard, the present study was conducted to investigate serum homocysteine level and its relationship with internal carotid intima thickness in ESKD patients undergoing routine hemodialysis.
Methods. This study comprised 56 ESKD patients, older than 40 years, undergoing hemodialysis for at least 1 year. All participants were taking Nephrovit for at least 6 months. The study participants were patients who underwent ultrasonography for CIMT determination and laboratory test
Results. There was no statistically significant relationship between the mean homocysteine level and hypertension, diabetes mellitus, duration of dialysis, and body mass index (BMI). Among the study participants, the results also showed that the mean value of CMIT homocysteine and C-reactive protein (CRP) were 0.89 millimeters, 30.44 (mcmol/L), and 35.60 mg/L; respectively. Despite hypertension, there was a significant difference between the mean values of CMIT in patients with diabetes mellitus and those who had been on dialysis for a longer period (more than 3 years). Also, the mean value of CMIT was significantly higher in obese patients than those with normal BMI. None of the other variables including homocysteine serum level, C-reactive protein (CRP), and CMIT showed a significant correlation.
Conclusion. The results of the study suggest that there is no relationship between serum homocysteine level and carotid intima-media thickness in hemodialysis patients.

IJKD 2023;17:222-7
www.ijkd.org
DOI: 10.52547/ijkd.7424

INTRODUCTION

Homocysteine is an intermediate amino acid formed during the conversion of methionine to cysteine, and its elevation in serum has been reported to be associated with atherosclerotic vascular disease and venous thromboembolism.

Elevated plasma homocysteine levels can be both hereditary and acquired. The acquired form is mostly due to folic acid, vitamin B6 or vitamin B12 deficiency, chronic kidney disease, medications (fibrates, nicotinic acid, methotrexate, metformin), and smoking.[1]

Homocysteine-induced vascular damage includes increased intimal thickening, disruption of the elastic lamina, smooth muscle hypertrophy, and platelet aggregation which lead to narrowing of the lumen and downstream ischemia.[2]

In CKD patients, hyperhomocysteinemia may be caused by several factors, including impaired renal function, decreased homocysteine metabolism, and deficiencies of vitamins B6, B12, and folate. Treatment options for hyperhomocysteinemia in CKD patients include vitamin supplementation, dietary alterations, and use of medications that lower homocysteine levels.[3]

Generally, we measure serum homocysteine only in patients with suspected homocystinuria but not in other clinical conditions because measures for lowering homocysteine levels have not been shown to prevent cardiovascular disease or venous thromboembolism.[4] In addition, no statistically significant decrease in recurrent thromboembolism or cardiovascular disease was observed in asymptomatic patients treated with B vitamins to lower homocysteine levels.[5]

Early detection and treatment of hyperhomocysteinemia may be crucial in improving outcomes in CKD patients. Further research is needed for better understanding of the mechanisms underlying this condition in CKD and developing more effective treatment strategies.[3]

As mentioned in previous studies, according to study done by Lubomirova *et al.* in relation of homocysteine and carotid atherosclerosis in chronic renal failure and other studies,[6,7] homocysteine increases arterial intimal thickness, and in this study, we investigated the relationship between homocysteine levels in ESKD patients and CIMT, which is an indicator of future coronary artery disease (CAD) and stroke.[8]

MATERIALS AND METHODS

Patients and Settings

Fifty-six ESKD patients who received hemodialysis for at least 1 year at Shahid Beheshti, Ayatollah Yathrabi, and Seyed Al-Shohdai hospitals in Aran and Bidgol City, from January 2019 to September 2022, were enrolled in this cross-sectional study.

The inclusion criteria were:

1. suffering from stage 5 CKD (ESKD).
2. undergoing routine hemodialysis, 12 hours per week, for at least one year, who have a KT/V > 1.2.
3. having age 40 years and older
4. receiving 500 micrograms of folic acid, 40 mg of vitamin B6, and 1000 micrograms of vitamin B12 supplements for 6 months.
5. having Body mass index (BMI) between 18.5 and 30.

The exclusion criteria were:

1. Cigarette smoking or consuming serum homocysteine levels increasing medications.
2. suffering from acute illness, hospitalization, or surgery in the past 6 months.
3. having hereditary hyperhomocysteinemia (homocysteine over 100 mcmol/L).
4. not taking Nephrovit supplement properly.

Demographic profiles were obtained from the patients through interviews and medical records.

Assessment of CIMT

For measurement of CIMT, all patients underwent ultrasonography for end-diastolic velocity estimation and spectral configuration in supine position by an experienced radiologist in vascular system Doppler ultrasound. The used instrument was a 7.5-MHz linear high-resolution fully automated Doppler ultrasound device (Samsung WS80A), capable of detecting changes in arterial wall thickness of 0.1 mm accuracy. The carotid arteries were examined bilaterally, considering the proximal part of the internal and external carotid arteries, with longitudinal and transverse images. All measurements were taken 2 cm proximal to the carotid bulb near the arterial wall. A CIMT above the 75th percentile of the mean for age, sex, and ethnicity or an absolute thickness greater than 1.0 mm was considered abnormal.[9]

Homocysteine Measurement

Homocysteine levels in the serum were measured after overnight fasting, by a simple blood test and the results were reported in micromoles per liter (µmol/L).

Other factors such as diet, smoking, and stress before blood sampling was also considered. In borderline cases, we repeated the test at least twice to confirm elevated levels before making a diagnosis of hyperhomocysteinemia.

Statistical Analysis

All statistical analyses were calculated by SPSS for Windows, version 17.0, The Kolmogorov-Smirnov

test was used to test whether the data followed a normal distribution. Quantitative variables such as age, BMI, serum homocysteine and CRP levels, duration of dialysis treatment, and CIMT were reported as mean and standard deviation or range. For qualitative variables, frequency and percentage were calculated. Pearson correlation coefficient, t-test, or ANOVA were calculated for parametric variables, and Mann-Whitney or Spearman correlation test for nonparametric variables. $P < 0.05$ was considered statistically significant.

RESULTS

This study aimed to evaluate the relationship between serum homocysteine level and CIMT in hemodialysis patients. Fifty-six ESKD patients on routine hemodialysis were enrolled in this study, out of which 32 (57.1%) were male and 24 (42.9%) were female. The prevalence of diabetes mellitus and hypertension in the study population were 43 (76.8%) and 27 (48.2%), respectively. The descriptive indicators of variables including age, duration of dialysis, blood homocysteine level, CRP, BMI, and CMIT are shown in Table 1.

The association between the study variables indicate that there was no statistically significant difference between the mean serum homocysteine levels and hypertension, diabetes mellitus, hemodialysis duration, and BMI in the study participant (Table 2).

The relationship between the mean CMIT and hypertension, diabetes mellitus, hemodialysis duration, and BMI in the study participants are shown in Table 3. Data analysis using T-test showed that there was no statistically significant difference between the mean CMIT in terms of hypertension; however, the difference between the mean CMIT in terms of diabetes mellitus, hemodialysis duration, and BMI was statistically significant.

The mean CMIT was significantly higher in patients with history of diabetes mellitus, those who underwent dialysis for a longer period of time, and overweight individuals.

The results of the study on correlation of study variables, using the Pearson test are shown in Table 4. No significant correlation was found between the study variables (e.g., homocysteine, CRP, and CMIT).

Table 2. Mean Serum Homocysteine Level in Terms of Hypertension, Diabetes Mellitus, Hemodialysis Duration, and BMI

Variable	Mean	Standard Deviation	*P*
Hypertension (BP > 140/90)			
Yes	29.92	2.40	> .05
No	30.93	2.53	
Diabetes mellitus			
Yes	30.62	2.43	> .05
No	29.84	2.73	
Hemodialysis duration, y			
1-5	30.34	2.62	> .05
>5	30.66	2.24	
BMI, kg/m^2			
18.5 to 24.9	30.31	2.33	> .05
25 to 29	31.00	3.16	

Table 3. Mean CMIT in Terms of Hypertension, Diabetes Mellitus, Hemodialysis Duration, and BMI

Variable	Mean	Standard Deviation	*P*
Hypertension (BP > 140/90)			
Yes	0.893	0.352	> .05
No	0.897	0.335	
Diabetes mellitus			
Yes	0.947	0.364	< .05
No	0.723	0.164	
Hemodialysis duration, y			
1-5	0.805	0.266	< .05
>5	1.08	0.407	
BMI, kg/m^2			
18.5 to 24.9	0.636	0.345	< .05
25 to 29	0.958	0.150	

DISCUSSION

Hyperhomocysteinemia is a condition

Table 1. Descriptive Indicators of Variables: Age, Hemodialysis Duration, Serum Homocysteine Level, CRP, BMI, CMIT

Variable	Min	Max	Mean	Standard Deviation
Age, y	42	81	58.07	8.15
Hemodialysis duration, y	3	12	5.78	1.72
Homocysteine level, µmol/L	25	35	30.44	2.50
CRP, mg/L	6	68	35.60	16.56
BMI, kg/m^2	19	26	23.03	1.65
CMIT, mm	0.4	2.2	0.89	0.34

Table 4. Correlation Between Homocysteine, CRP, and CMIT

Variable		Homocysteine	CRP	CMIT
Homocysteine	Coefficient		-0.240	-0.068
	P value		.075	.621
CRP	Coefficient	0.240		0.224
	P value	.075		.096
CMIT	Coefficient	-0.068	0.224	
	P value	.096	.621	

characterized by increased serum levels of homocysteine, an amino acid that is common in patients with chronic kidney disease (CKD) and is associated with an increased risk of cardiovascular diseases, kidney failure, and mortality.[10]

In CKD patients, hyperhomocysteinemia can be caused by several factors, including impaired kidney function, decreased homocysteine metabolism, and vitamins B6, B12, and folate deficiencies. Treatment options for hyperhomocysteinemia in CKD patients include vitamin supplementation, dietary modification, and use of medications that lower serum homocysteine levels.[3]

Early detection and treatment of hyperhomocysteinemia may be crucial in reducing the risk of cardiovascular events and slowing the progression of kidney disease in CKD patients.[3]

Studies have shown that elevated serum homocysteine levels may be associated with an increased risk of atherosclerosis and cardiovascular diseases. Specifically, high serum homocysteine levels have been associated with an increase in CIMT.[11] In individuals with hyperhomocysteinemia, CIMT levels are significantly higher than those with normal homocysteine serum levels. The study by Liu *et al.*, also showed that treatment with folic acid and vitamin B12, which are known to lower serum homocysteine levels, resulted in a significant reduction in CIMT.[11]

Currently, it has been reported that atherosclerotic changes in the CIMT, measured by ultrasound, reflect atherosclerosis of the coronary arteries and cerebrovascular diseases.[12]

The study by Taruangsri *et al.* showed that older age, male sex, longer duration of hemodialysis, and higher serum homocysteine level were related to atherosclerosis in univariate analysis, but in multivariate analysis, serum homocysteine level showed no significant association with atherosclerosis and CIMT,[13] which is concordant with the findings of our study.

Normal intima-media thickness of the carotid artery is usually between 0.6 and 0.7 mm in healthy middle-aged adults. Test results showing a thickness greater than 1.0 mm may indicate the risk of cardiovascular disease.[14]

The results of our study showed that the average CMIT value in the study participants was 0.89. In the studies conducted in ESKD patients, no specific cut-off value was established for determination of normal CIMT value; therefore, international studies indicated different values. For instance, a study in Japan found a CIMT value of 0.66, whereas another study in Turkey found a CIMT value of 0.99 in patients receiving hemodialysis.[15,16] Therefore the CIMT value in our study was compared with the cut-off value of the Turkish study and determined to be greater than normal when compared to the Japanese study.

Borràs M *et al.* showed that hemodialysis patients have greater global CIMT than patients receiving peritoneal dialysis that is an independent factor associated with lower CIMT.[17]

In a study conducted in India, CMIT was found to increase in the group of conservatively treated stage 4 CKD patients and in the group of stage 5 patients undergoing hemodialysis four times per week compared to the group with stage 3 CKD.[13]

Sánchez-Álvarez *et al.* showed that mean intima-media thickness (CIMT) was higher in patients on maintenance hemodialysis than in the control group (0.947 ± 0.308 vs. 0.619 ± 0.176 mm, $P < .001$). Intima-media thickness was associated with age (r = 0.268, $P = .038$), diabetes mellitus (r = 0.650, $P < .001$), and hypertension (r = 0.333, $P = .012$), but not with serum homocysteine levels.[18]

Some authors have linked the occurrence of cardiovascular events with the sum of increased inflammatory markers such as CRP, which are significantly elevated in hemodialysis patients compared with the normal population.[19,20] The results of our study showed that the mean value of CRP was 35.60 mg/L, which was above the

normal range of normal individuals.

The results of our study also showed that the mean value of serum homocysteine was 30.44 (mcmol/L) which was concordant with the findings of Jain *et al.* who found a statistically significant difference between homocysteine levels in three groups of CKD patients (stages 3, 4, and 5).[21]

The results of our study showed that none of the variables such as diabetes mellitus, hypertension, dialysis duration, and BMI affected the mean serum homocysteine levels, while diabetes mellitus, dialysis duration, and BMI affected CMIT, such that the mean CMIT value was significantly higher in patients with diabetes mellitus, those with longer dialysis duration, and overweight patients (BMI over 25). Our study also did not find a significant correlation between serum homocysteine levels and CMIT. Previous studies on carotid ultrasound measurements in hemodialysis patients have shown that CMIT value is linked to atherosclerosis and serum homocysteine levels, as in the normal population.[22]

Despite traditional atherosclerosis risk factors, hyperlipidemia, hyperparathyroidism, greater high sensitivity CRP (hs-CRP) levels, lower serum albumin, hyperphosphatemia, and higher calcium-phosphate product are common risk factors for the development of atheroma and thickening of arterial walls in hemodialysis patients, which should be considered as variables in future studies to determine the exact role of serum homocysteine in CMIT.[23-25]

There are conflicting reports regarding the relationship between obesity and kidney disease, with some studies suggesting that adipose tissue may have a paradoxical protective effect. This may be due to the production of hormones by adipose tissue that have anti-inflammatory properties and improve insulin sensitivity. In addition, overweight or obese individuals may have more muscular mass, which is associated with better kidney disease outcomes.[26] The study by Kalantar-Zadeh *et al.* showed that even traditional risk factors have a different effect in arterial disease.[26]

Thus, it is evident that a potential contradiction between our study and other studies that indicate a strong positive correlation between serum homocysteine levels and CIMT, may be due to demographic variables such as age that must be considered in ESKD and healthy individuals.

Studies have shown that there is an association between hyperhomocysteinemia and CIMT. However, it is not clear whether hyperhomocysteinemia directly causes increased CMIT or it is simply a marker of other underlying conditions that contribute to elevated CMIT.[27-30]

CONCLUSION

The study findings suggested no association between serum homocysteine levels and carotid intima-media thickness (CIMT)in hemodialysis patients. Furthermore, none of the other variables including dialysis duration, BMI, diabetes mellitus, and hypertension had an impact on serum homocysteine levels. However, diabetes mellitus, longer dialysis time, and obesity (BMI greater than 25) affected the average CIMT in hemodialysis patients.

To reduce the risk of cardiovascular diseases, it is important to manage hyperhomocysteinemia through lifestyle modifications (e.g., diet and exercise) and/or medications. In hyperhomocysteinemia, regular monitoring of CMIT may also be recommended to detect changes in arterial thickness and to take the necessary measures.

DECLARATION

Funding

This research received no grant from any funding agency.

Conflicts of Interest / Competing Interests

The authors declare no conflict of interest.

Ethics Approval

In this study, no additional costs were imposed on the patients. We maintained the patients' privacy, and all participants signed a written consent. This study was approved by the ethic committee of Kashan university of medical science with the ethical code: IR.KAUMS.MEDNT.REC.1399.12.2

Availability of Data and Material

The data that support the findings of this study are available from the corresponding author, [SH.T], upon reasonable request.

Authors' Contributions

SH.T., and A.S. contributed to data collection, writing, drafting, and critical appraisal of the manuscript. M.K, F.GH, SZ.T, M.S, ST.S . contributed to scientific writing and final revisions, and H.M. contributed to data analysis.

REFERENCES

1. Desouza, C., et al., Drugs affecting homocysteine metabolism. Drugs, 2002. 62(4): p. 605-616.
2. Lubomirova, M., et al., Homocystein and carotid atherosclerosis in chronic renal failure. Hippokratia, 2007. 11(4): p. 205.
3. Badri, S., et al., Homocysteine-Lowering Interventions in Chronic Kidney Disease. J Res Pharm Pract, 2021. 10(3): p. 114-124.
4. Undas, A., J. Brozek, and A. Szczeklik, Homocysteine and thrombosis: from basic science to clinical evidence. Thromb Haemost, 2005. 94(5): p. 907-15.
5. Stanger, O., et al., Clinical use and rational management of homocysteine, folic acid, and B vitamins in cardiovascular and thrombotic diseases. Z Kardiol, 2004. 93(6): p. 439-53.
6. Lubomirova, M., et al., Homocystein and carotid atherosclerosis in chronic renal failure. Hippokratia, 2007. 11(4): p. 205-9.
7. Ntaios, G., et al., Homocysteine and carotid intima-media thickness in ischemic stroke patients are not correlated. Neuropsychiatr Dis Treat, 2008. 4(2): p. 477-9.
8. Kang, S.-S., P.W. Wong, and M.R. Malinow, Hyperhomocyst (e) inemia as a risk factor for occlusive vascular disease. Annual review of nutrition, 1992. 12(1): p. 279-298.
9. Sheikhy, A., et al., Personalized medicine in cardiovascular disease: review of literature. Journal of Diabetes & Metabolic Disorders, 2021. 20(2): p. 1793-1805.
10. van Guldener, C. and K. Robinson, Homocysteine and renal disease. Semin Thromb Hemost, 2000. 26(3): p. 313-24.
11. Liu, C.P., et al., The Impact of Metabolic Syndrome, Homocysteine, and B Vitamins on Carotid Artery Intima-Media Thickness in Hypertensive Patients. Acta Cardiol Sin, 2013. 29(1): p. 56-63.
12. Ray, J., Heart Outcomes Prevention Evaluation 2 (HOPE-2) Investigators. Homocysteine-lowering therapy and risk for venous thromboembolism: a randomized trial. Ann Intern Med., 2007. 146: p. 761-767.
13. Taruangsri, P., et al., Relationship between hyperhomocysteinemia and atherosclerosis in chronic hemodialysis patients. JOURNAL-MEDICAL ASSOCIATION OF THAILAND, 2005. 88(10): p. 1373.
14. Kasliwal, R.R., et al., Carotid intima-media thickness: Current evidence, practices, and Indian experience. Indian J Endocrinol Metab, 2014. 18(1): p. 13-22.
15. Ishizaka, N., et al., Association between chronic kidney disease and carotid intima-media thickening in individuals with hypertension and impaired glucose metabolism. Hypertens Res, 2007. 30(11): p. 1035-41.
16. Mutluay, R., et al., Dialysis type may predict carotid intima media thickness and plaque presence in end-stage renal disease patients. Adv Ther, 2012. 29(4): p. 370-82.
17. Borràs, M., et al., Peritoneal Dialysis Is an Independent Factor Associated to Lower Intima Media Thickness in Dialysis Patients Free From Previous Cardiovascular Disease. Front Physiol, 2018. 9: p. 1743.
18. Sánchez-Álvarez, J.E., et al., Carotid ultrasound: prevention of heart disease and mortality on haemodialysis. Nefrología (English Edition), 2010. 30(4): p. 427-434.
19. Sabzevari, E., K. Shahanipour, and A. Moshtaghie, The Levels of Inflammatory Markers (IL-6, CRP), Homocysteine and Lipid Profile in Hemodialysis Patients, Isfahan. Hormozgan Medical Journal, 2019. 23(3).
20. Holland, Z., et al., Carotid intima-media thickness is a predictor of coronary artery disease in South African black patients. Cardiovasc J Afr, 2009. 20(4): p. 237-9.
21. Jain, D., et al., Evaluation of serum homocysteine level and its relation with carotid intima-media thickness in patients of chronic kidney disease. Medical Studies/Studia Medyczne, 2017. 33(4): p. 247-253.
22. Arzpeyma, S.F., et al., Studying the relationship between the risk factors of cardiovascular diseases and carotid intima-media thickness (CIMT) in renal patients undergoing hemodialysis admitted to Razi Hospital and Caspian Dialysis Center in Rasht, 2017. Revista Latinoamericana de Hipertensión, 2018. 13(3): p. 251-258.
23. Nakashima, A., et al., Different risk factors for the maximum and the mean carotid intima-media thickness in hemodialysis patients. Internal medicine, 2003. 42(11): p. 1095-1099.
24. Yadollah-Damavandi, S., et al., Topical Hypericum perforatum improves tissue regeneration in full-thickness excisional wounds in diabetic rat model. Evidence-Based Complementary and Alternative Medicine, 2015. 2015.
25. Moosaie, F., et al., Effects of Pentoxifylline on Serum Markers of Diabetic Nephropathy in Type 2 Diabetes. Diabetes Therapy, 2022. 13(5): p. 1023-1036.
26. Kalantar-Zadeh, K., et al., The Obesity Paradox in Kidney Disease: How to Reconcile it with Obesity Management. Kidney Int Rep, 2017. 2(2): p. 271-281.
27. Hojs, R., Carotid intima-media thickness and plaques in hemodialysis patients. Artificial organs, 2000. 24(9): p. 691-695.
28. Suliman, M.E., et al., Plasma pentosidine and total homocysteine levels in relation to change in common carotid intima-media area in the first year of dialysis therapy. Clinical nephrology, 2006. 66(6): p. 418-425.
29. Ossareh, S., A. Alaei, and D. Saedi, Carotid intima-media thickness in maintenance hemodialysis patients: role of cardiovascular risk factor. Iran J Kidney Dis, 2011. 5(3): p. 169-74.
30. Ekart, R., et al., Predictive value of carotid intima media thickness in hemodialysis patients. Artif Organs, 2005. 29(8): p. 615-9.

Correspondence to:
Seyed Hamed Tabatabaei
Kashan University of Medical Sciences, Kashan, Iran
ORCID ID: 0000-0002-0425-6973
E-mail: Dr.tabatabaei67@gmail.com

Received February 2023
Revised April 2023
Accepted June 2023