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NEPHROLOGY - ORIGINAL PAPER

The factors that affect plasma homocysteine levels, pulse wave velocity and their relationship with cardiovascular disease indicators in peritoneal dialysis patients

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Abstract
Objective The incidence of cardiovascular diseases (CVD) is high in end-stage renal disease (ESRD) population and hyperhomocysteinemia is an important CVD risk factor. The aims of this study are to asses the incidence of hyperhomocysteinemia and the factors that affect the homocysteine (Hcy) levels in peritoneal dialysis (PD) patients, and to analyze the relationships between Hcy levels and clinical and echocardiographic CVD, and the pulse wave velocity (PWV).
Study design Sixty ESRD patients undergoing PD for at least 6 months were included in the study. Biochemical parameters, echocardiography, and PWV were analysed for every subject.
Results Mean Hcy level was 27.2 ± 15.7 μmol/L and was high in 53 patients (88.3%). Fibrinogen, dialysate/plasma creatinine ratio and folic acid were found to be the independent predictors of Hcy level ($P < 0.001$; $P < 0.01$; $P < 0.05$, respectively). Patients with atherosclerosis had significantly higher plasma Hcy levels ($P < 0.05$). No significant relationship was found between plasma Hcy levels and echocardiographic findings and PWV.
Conclusion Hyperhomocysteinemia incidence seems high among PD patients and despite significant relationship between fibrinogen and Hcy in our study, it is essential to evaluate the link between Hcy levels and inflammation. Folic acid replacement even in normal folic acid levels, enough elimination of volume and solutes may be beneficial to control Hcy levels, whereas PWV was found to be related with comorbidities and and dialysate kinetics.

Keywords End stage renal disease · Peritoneal dialysis · Homocysteine · Atheroclerotic heart disease · Echocardiography · Pulse wave velocity

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Introduction

Despite advances in dialysis, pharmacology, and patient rehabilitation programs, mortality and morbidity rates of end-stage renal disease (ESRD) are still high and cardiovascular diseases (CVD) constitute more than half of them [1, 2]. Besides the classical cardiovascular risk factors such as diabetes, hypertension, and smoking, there are additional risk factors like volume overload, proteinuria, high

plasma uremic toxin levels, inflammation, abnormal calcium-phosphorus metabolism, and hyperhomocysteinemia in this patient group [3].

Homocysteine (Hcy) is a sulfur-containing amino-acid whose high levels were shown to be a cardiovascular risk factor in general population [4, 5]. Hcy causes oxidative damage to vascular endothelium and results in vascular smooth muscle proliferation and atherogenesis, and increases tendency to thrombotic events [6]. Hyperhomocysteinemia incidence was found to be high in hemodialysis (HD) and peritoneal dialysis (PD) patients [7, 8]. The exact mechanism of this increase is not clear and the decreased renal clearence of Hcy is not enough to explain it [8, 9]. Vitamin deficiencies, genetic polymorphisms of enzymes involving Hcy metabolism and the effects of uremic toxins on degradation of Hcy may be possible causes [10–12].

Arterial system is affected by ESRD-related risk factors and traditional risk factors in this patient group, as shown in studies evaluating arterial intimal and medial calcification and thickening of carotid arteries by noninvasive methods [13, 14]. Pulse wave velocity (PWV) is another indirect measure of arterial stiffness [15]. The velocity of the pulse wave depends on the features of arterial wall, which are wall elasticity and radius of the vessel, according to Moens-Korteweg equation [16]. The pressure increases in systole and decreases in diastole. The pulse waves and reflected waves are analyzed as closed system of wave models of Windkessel, as computerized analyses for noninvasive applications [16, 17]. The measurements are generally performed between femoral, carotid, radial, brachial or dorsalis pedis pulses [16].

In this study, hyperhomocysteinemia rate in chronic PD patients was evaluated. The factors that affect the plasma Hcy levels and the relationships between Hcy levels and the echocardiographic findings and arterial stiffness, which is evaluated by PWV, were also analysed.

Materials and methods

Sixty ESRD patients [34 women (56.7%) and 26 men (43.3%)] who are followed-up in our outpatient PD Unit for at least 6 months were included in the study. Mean age was 46.2 ± 13.2 years (range: 20–69 years) and median follow-up period was 42.5 months (6–108 months). Forty-seven patients were having continuous ambulatory peritoneal dialysis (CAPD) and 13 patients were having automated peritoneal dialysis (APD). Patients who have a history of chronic infection, paranchymal liver disease, hypothyroidism, malignancy and drugs which may affect plasma Hcy levels were excluded. All patients gave their informed consents.

Mean of at least two measurements of plasma Hcy levels and demographic features, comorbid diseases, cardiovascular events, blood biochemistry, blood pressure measurements, PD schedule, residual renal function (RRF), and data about kinetic tests for permeability of peritoneal membrane, adequacy of PD and nutritional status were recorded. Smoking status and history of medications (including erythropoietin doses calculated for each week and per kilo of the patient) were also recorded. Measurements during a systemic or PD-related infection were not included.

Body mass index (BMI) was calculated by [weight(kg)/height $(cm)^2$] formula and the body surface area was calculated from Du Bois nomograms [18]. Charlson comorbidity index was used for comorbidity scoring [19].

Patients with systolic blood pressure exceeding 140 mmHg and diastolic blood pressure exceeding 90 mmHg and patients who were normotensive with antihypertensive drugs were accepted as hypertensive. Means of systolic and diastolic blood pressures and pulse pressures were recorded.

Plasma Hcy levels were analysed with high-pressure lipid chromatography (HPLC) and levels exceeding 14 μmol/L were accepted as hyperhomocysteinemia. The values between 15 and 30 μmol/L were mild, ≥30 μmol/L were severe and ≥100 μmol/L were very severe hyperhomocysteinemia according to medical literature [20, 21]. Parathormone, vitamin B12 and folic acid levels were measured by radioimmunoassay (SimulTRAC-SNB), and lipoprotein (a) levels by turbidimetric immunoassay. CRP, prealbumin levels were evaluated by nephelometric immünoassay (Dade-Behring BN II), while glucose, albumin, total cholesterol, triglyceride, LDL- and HDL-cholesterol levels were measured by Beckman LX 20 autoanalyzer.

Permeability of the peritoneal membrane was evaluated with standard 4 hour-peritoneal equilibration test (PET). D/P creatinin ratio (D/PCrea),

residual renal function, normalized urea clearance (Kt/Vurea), and normalized protein equivalent of nitrogen appearance (nPNA) were calculated.

Echocardiographic evaluation and arterial stiffness measurements were performed at the end of the follow-up. Echocardiography was performed by the same experienced clinician. Left ventricular mass (LVM) and left ventricular mass index (LVMI) were calculated according to Devereux's formula [22].

Pulse-wave velocity (PWV) measurements were performed with SphygmoCor® Pulse Wave Velocity System by the same experienced technician. The height, body weight, arterial blood pressure values, age, and sex of all patients were recorded and patients underwent PWV measurement. The distances between supraclavicular notch and radial and femoral pulses were recorded. The difference between these two distances were recorded as the way that pulse wave passes. Then pulse wave was recorded using three electrocardiograph electrodes on their trunk. PWV recordings were analyzed by software system of the SphygmoCor®, and the results were recorded as velocity (m/s) ± SD.

Results were defined as mean ± SD for continuous variables, and as percentages for categoric variables. Student's T, ANOVA, Mann–Whitney U and Kruskal Wallis tests were used for comparison of group means. The relationships between variables were analyzed by using Spearman rank correlation coefficient. Stepwise multiple regression analyses were performed to evaluate the factors independently affecting plasma Hcy levels and PWV. Differences were considered significant when $P < 0.05$.

Results

Mean plasma Hcy level was 27.2 ± 15.7 μmol/L and 53 patients (88.3%) had hyperhomocysteinemia. Hcy was higher than 30 μmol/L in 17 patients (28.3%). Men had insignificantly higher Hcy levels than women. Hcy levels did not differ by to the smoking status, ethiology of renal failure and medications. Hypertensive and normotensive patients had similar Hcy levels. Fibrinogen showed a positive correlation with plasma Hcy, and there were significantly negative correlations between Hcy levels and vitamin B12 and folic acid levels ($r = 0.502$, $P < 0.001$; $r = -0.257$, $P < 0.05$; $r = -0.306$, $P < 0.05$, respectively) (Figs. 1, 2, 3). Plasma creatinine was the only biochemical parameter that had correlation with plasma Hcy ($r = 0.328$, $P < 0.01$). Hcy showed a negative correlation with Kt/Vurea and total fluid intake and a positive correlation with D/PCrea ($r = -0.270$, $P < 0.05$; $r = -0.277$, $P < 0.05$; $r = 0.374$, $P < 0.01$, respectively). Hcy levels were significantly higher in patients with high peritoneal permeability ($n = 6$) when compared with the patients with low permeability (43.8 ± 28.3 vs. 25.6 ± 12.7 μmol/L, $P < 0.01$) (Fig. 4). Hcy levels did not differ between CAPD and APD patients (25.6 ± 12.7 μmol/L vs. 33.1 ± 23.2 μmol/L, $P > 0.05$). In stepwise multiple linear regression analysis fibrinogen, folic acid, and D/P Creatinine ratio were found to be independent variables for plasma Hcy levels. Although fibrinogen was related with Hcy levels, relationship with albumin and CRP was not statistically significant in this analysis ($P > 0.05$) (Table 1).

Thirteen patients (25%) had coronary artery disease and four patients (6.7%) had peripheric arterial disease. Patients with a history of atherosclerotic heart disease had significantly higher plasma Hcy levels when compared with the disease-free ones (34.0 ± 19.5 vs. 24.7 ± 13.4 μmol/L, $P < 0.05$). Patients with peripheral arterial disease had insignificantly higher Hcy levels when compared with the disease free ones.

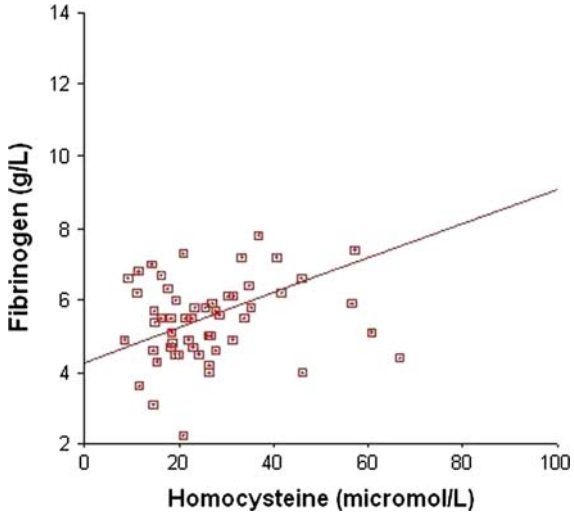

Fig. 1 The relationship between plasma Hcy and fibrinogen levels

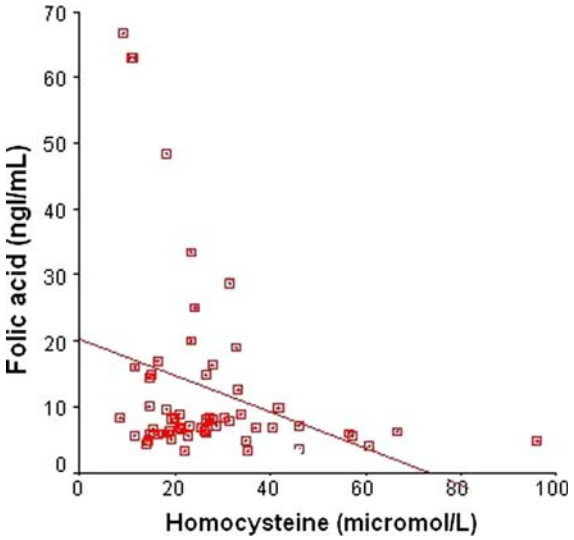

Fig. 2 The relationship between plasma Hcy and folic acid levels

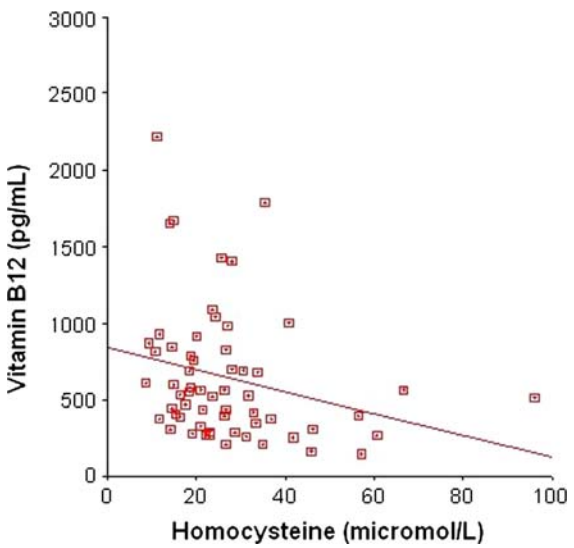

Fig. 3 The relationship between plasma Hcy and vitamin B12 levels

Echocardiographic evaluation was performed in 53 patients and 45 patients (84.9%) had left ventricular hypertrophy, and five patients (9.4%) had left ventricular systolic dysfunction. No significant relationship was found between echocardiographic findings and plasma Hcy levels.

A positive correlation between PWV and pulse pressure was found ($r = 0.359$, $P < 0.01$). PWV was significantly higher in patients who had atherosclerotic heart disease and peripheric arterial disease when compared with the disease free ones (10.1 ± 3.3 vs. 7.3 ± 2.0 m/sn; $P < 0.01$ and 11.4 ± 3.0 vs. 7.8 ± 2.5 m/sn; $P < 0.01$, respectively). Men had insignificantly higher values of PWV than women. Patients with CAPD had significantly higher PWV than APD patients (8.5 ± 2.6 vs. 6.7 ± 2.4 m/sn; $P < 0.05$). There were significant negative correlations between PWV and exchange number and total infusion volume ($r = -0.305$, $P < 0.05$; $r = -0.300$, $P < 0.05$, respectively). There was no relationship between PWV and dialysis schedule and parameters of dialysis adequacy.

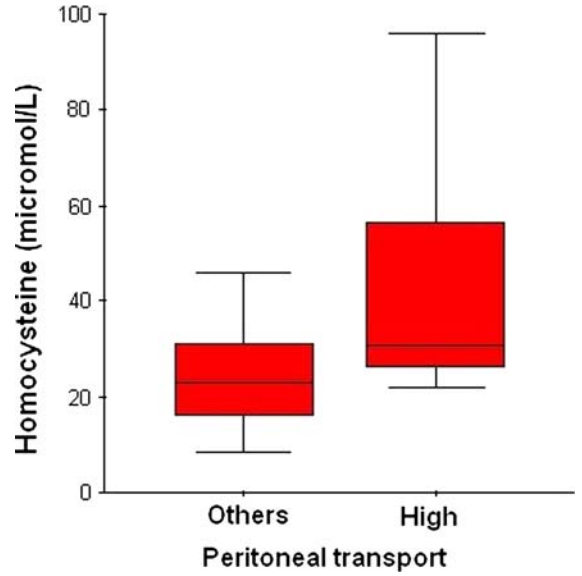

Fig. 4 The relationship between plasma Hcy and peritoneal permeability

Table 1 The independent variables affecting plasma Hcy levels (IS: Insignificant)

	Beta	*t*	*P*
Fibrinogen	0.457	4.153	<0.001
Dialysate/plasma creatinine	0.301	2.738	<0.01
Folic acid	−0.235	−2.168	<0.05
Creatinine	0.194	1.722	IS
Vitamin B_{12}	−0.155	−1.383	IS
Total fluid intake	−0.092	−0.662	IS
Kt/V_{urea}	−0.038	−0.243	IS
CRP	0.022	0.141	IS
Albumin	0.010	0.064	IS

Table 2 The independent variables affecting PWV (IS: Insignificant)

	Beta	*t*	*P*
Co-morbidity index	0.544	4.493	<0.001
Age	0.368	3.004	<0.01
Infusion volume	−0.242	2.136	<0.05
Diabetes mellitus	0.323	1.516	IS
Hemoglobin levels	0.213	1.376	IS
Hypertension	0.138	0.982	IS
Lipoprotein (a)	0.114	0.913	IS
Number of exchanges	0.182	0.847	IS
Calcium × phosphorus	0.108	0.688	IS
CAPD	0.083	0.438	IS
Weekly erythropoietin dose	−0.069	−0.431	IS
Glucose	0.037	0.184	IS

Significant relationships between PWV and comorbidity index, age, hemoglobin levels, weekly erythropoietin doses, lipoprotein(a), fasting plasma glucose, calcium × phosphorus product, presence of hypertension, and diabetes were found (Table 2). In stepwise multiple linear regression analysis, comorbidity index, age, and infusion volume were found to be the the independent variables related with PWV ($P < 0.001$; $P < 0.01$; $P < 0.05$, respectively). Aortic diameter, left atrium diameter, left ventricule end-diastolic diameter and left ventricular hypertrophy were found to have significant relationships with PWV ($r = 0.340$, $P < 0.05$; $r = 0.352$, $P < 0.05$; $r = 0.293$, $P < 0.05$, respectively).

Discussion

ESRD patients on maintenance dialysis have 10–100 times higher CVD prevalance when compared with normal population and CVD constitute more than half of the deaths in this population [1, 2]. Hyperhomocysteinemia has been shown to be an independent risk factor for CVD in general population and it is more frequent in ESRD patients [8, 22]. In the current study, the incidence of hyperhomocysteinemia in PD patients is assessed and the factors affecting plasma Hcy levels were analysed. The relationships between hyperhomocysteinemia and PD schedule, dialysis adequacy, biochemical parameters, echocardiographic findings, and PWV as an indicator of arterial stiffness and atherosclerosis were determined.

In our study, 88.3% of the patients showed high Hcy levels (>14 μmol/L), and in 28.3% it was higher than 30 μmol/L, which are concordant with the previous studies [19, 23]. We did not find values higher than 100 μmol/L like the previous studies. In several studies sex and age were found to be related with plasma Hcy levels, but we did not find any relationship with demographic features. In recent studies smoking was found to have an impact on Hcy levels in both sexes [24, 25]. Less number of smokers in our study may be the reason for our findings, which indicated no relationship between plasma Hcy and smoking.

Data about the relationship between hypertension and hyperhomocysteinemia are conflicting [26–28]. Hyperhomocysteinemia may have undesirable effects on endothelial functions that may cause the tendency to hypertension. Among 60 patients, 36 (60%) were hypertensive and we did not find any difference in Hcy levels between hypertensive and normotensive ones. There were no significant relationships between Hcy levels and diastolic and systolic blood pressures and pulse pressure.

Independent relationship between hyperhomocysteinemia and plasma fibrinogen that we have found in our study is similar with the study of Aso et al. may be a suggestion for the role of hyperhomocysteinemia on inflammation [29]. In vivo and in vitro studies show that Hcy may be a triggering factor for inflammation and the inflammation may be the mediator between hyperhomocysteinemia and vascular damage [30]. Conflicting reports in clinical studies and lack of significant relationship between CRP and albumin levels and Hcy in our study make it difficult to decide a definite conclusion about the relationship between Hcy and inflammation [31, 32]. Prospective studies with larger patient groups are needed to conclude about the relationship between inflammatory markers and Hcy levels.

Although none of our patients had folic acid deficiency, we observed that plasma folic acid level was an independent determinant for Hcy levels in PD patients. This finding may suggest the use of folic acid replacement in hyperhomocysteinemia in this patient group, even in cases of normal plasma folic acid levels. It has been shown to be beneficial in some recent reports, but its role in prevention of cardiovascular risks is still a controversy [33, 34].

It has been reported that total Hcy elimination is lower in APD patients and Hcy clearance is related with D/P Creatinine and total fluid drainage [35, 36]. In our study, plasma Hcy levels were insignificantly higher in APD patients when compared with CAPD. D/P Creatinine, Kt/Vurea and total fluid drainage were significantly related with Hcy levels, and only D/P Creatinine ratio was found to be an independent factor affecting Hcy levels. Based upon the knowledge of high peritoneal permeability as a poor prognostic factor in PD patients, we also observed higher Hcy levels in this patient group [37]. Besides hypervolemia due to insufficient fluid elimination and malnutrition due to excessive peritoneal protein loss, hyperhomocysteinemia may be another poor prognostic factor for these patients. Although the mechanism is still not clear and further studies are needed to clarify this, Vychytil et al. and Johnson et al. also suggested that hyperhomocysteinemia may be related with high peritoneal permeability [36, 38].

Hyperhomocysteinemia was found to be an important risk factor for both general population and ESRD patients [8, 23]. Hcy causes oxidative damage to vascular endothelium, which results with vascular smooth muscle proliferation and atherogenesis, and it also increases the tendency to thrombotic events [6, 39]. We observed higher Hcy levels in patients with CVD in our study. The increase in Hcy levels in patients with peripheral arterial disease was insignificant, but that may be probably due to less number of patients in this group. These findings that we have obtained about the relationship between atherosclerosis and Hcy are concordant with previous studies.

Previous studies explained the adverse effects of Hcy on endothelial functions with decreased biologic activity of nitric oxide [40]. Angiotensin converting enzyme inhibitors (ACEI) may cause increased nitric oxide release via bradykinin. ACEI and angiotensin-1 receptor blockers (ARB) can decrease asymmetric-dimethyl arginine (ADMA), which is a nitric oxide synthase inhibitor, and these drugs were thought to be relevant for patients with hyperhomocysteinemia [40]. But we did not find significant relationships between Hcy levels and ACEI or ARB medications in our study. This may be due to different medication periods among patients.

We evaluated the relationship between Hcy and echocardiographic findings in our patient group. Among them, 9% had left ventricular systolic dysfunction, but Hcy levels did not show any relationship with fractional shortening. In one study, a significant relationship between Hcy and left ventricular ejection fraction was observed in hypertensive patients but not in normotensives [41]. This inconcordant results may have several explanations; (1) Our patient sample may be lesser (2) Percentage fractional shortening values shows variability in very narrow scale in this patient group, (3) Our follow-up period may be too short to observe this relationship. We observed left ventricular hypertrophy in 85% of our patients, but there was no relationship between Hcy levels and presence of left ventricular hypertrophy. This latter issue needs to be evaluated in larger prospective studies with longer follow-up periods.

PWV is an important and independent factor which predicts cardiovascular events [25, 27, 42]. We found that patients with CAPD had significantly higher PWV than APD patients and this difference seems to be related with higher exchange number and infusion volumes in APD when compared with CAPD. In our study comorbidity index, age, and dialysate infusion volume were found as independent determinants of PWV. Existence of hypertension and diabetes were evaluated in comorbidity index and they are also significantly related with PWV.

The effect of Hcy on PWV was not evaluated in detail before. Blacher et al. showed the relationship between lower extremity PWV and Hcy levels in ESRD patients [43]. In another report PWV and Hcy levels were found to be correlated [44]. In the latter study patients with Hcy > 25 μmol/L had higher PWV than the lower quantile. There are several studies showing significant relationships in hypertensive patients but not in healthy people [45, 46]. We also did not observe any effect of plasma Hcy levels on PWV and we conclude that the short follow-up period and less number of patients in our study may be the reasons for our findings.

In conclusion, we report a high incidence of hyperhomocysteinemia in PD patients. Besides folic acid replacement even in normal folic acid levels, providing enough elimination of volume and solutes may be beneficial to control Hcy levels. Independent relationship between Hcy and fibrinogen levels that we have found in our study make us to suggest the possible role of inflammation together with other mechanisms of vascular damage in hyperhomocysteinemia or a role of inflammation in Hcy increase.

This relationship should be evaluated in detail in further studies. Our short follow-up period and less number of patients may be the reason for our finding of no relationship between Hcy levels, echocardiographic findings, and also with PWV. Prospective studies with larger series and longer follow-up periods might provide further data about these relationships in PD patients.

References

1. Levin A (2003) Clinical epidemiology of cardiovascular disease in chronic kidney disease prior to dialysis. Semin Dial 16:101–105
2. Sarnak MJ, Levey AS (1999) Epidemiology of cardiac disease in dialysis patients. Semin Dial 12:69–76
3. Remppis A, Ritz E (2008) Cardiac problems in the dialysis patient: beyond coronary disease. Semin Dial 21:319–325
4. Herrmann W, Herrmann M, Obeid R (2007) Hyperhomocysteinaemia: a critical review of old and new aspects. Curr Drug Metab 8:17–31
5. Clarke R, Daly L, Robinson K et al (1991) Hyperhomocysteinemia: an independent risk factor for vascular disease. N Engl J Med 324:1149–1155
6. Hankey GJ, Eikelboom JW (1999) Homocysteine and vascular disease. Lancet 354:407–413
7. Hultberg B, Andersson A, Sterner G (1993) Plasma homocysteine in renal failure. Clin Nephrol 40:230–234
8. Moustapha A, Gupta A, Robinson K et al (1999) Prevalance and determinants of hyper-homocysteinemia in hemodialysis and peritoneal dialysis. Kidney Int 55:1470–1475
9. Chauveau P, Chadefaux B, Coude M et al (1993) Hyperhomocysteinemia, a risk factor for atherosclerosis in chronic uremic patients. Kidney Int Suppl 41:S72–S77
10. Robinson K, Gupta A, Dennis V et al (1996) Hyperhomocysteinemia confers an independent increased risk of atherosclerosis in end-stage renal disease and is closely linked to plasma folate and pyridoxine concentrations. Circulation 94:2743–2748
11. Fodinger M, Buchmayer H, Heinz G et al (2001) Association of two MTHFR polymorphisms with total homcysteine plasma levels in dialysis patients. Am J Kidney Dis 38:77–84
12. Morimoto K, Haneda T, Okamoto K, Ishida H, Kikuchi K (2002) Methylene-tetrahydrofolate reductase gene polymorphism, hyperhomocysteinemia, and cardiovascular diseases in chronic hemodialysis patients. Nephon 90:43–50
13. Gelev S, Spasovski G, Dzikova S, Trajkovski Z, Damjanovski G, Amitov V, Sikole A (2008) Vascular calcification and atherosclerosis in hemodialysis patients: what can we learn from the routine clinical practice? Int Urol Nephrol 40:763–770 (Epub 2008 Jun 27)
14. Kocak H, Gumuslu S, Sahin E et al (2009) Relationship between carotid artery intima-media thickness and brachial artery flow-mediated dilation in peritoneal dialysis patients. Int Urol Nephrol 41:409–416 (Epub 2008 Dec 30)
15. Blacher J, Guerin AP, Pannier B, Marchais SJ, Safar ME, London GM (1999) Impact of aortic stiffness on survival in end-stage renal disease. Circulation 99:2434–2439
16. Cohn JN, Quyyumi AA, Holberg NK, Jamerson KA (2004) Surrogate markers for cardiovascular disease: functional markers. Circulation 109(25 Suppl 1):IV31–IV46
17. London GM, Marchais SJ, Safar ME, Genest AP, Metivier F, Chedid K, London AM (1990) Aortic and large artery compliance in end-stage renal disease. Kidney Int 37: 137–142
18. Du Bois D, Du Bois EF (1989) A formula to estimate the approximate surface area if height and weight be known.1916. Nutrition 5:303–311
19. Charlson ME, Pompei P, Ales KL, MacKenzie CR (1987) A new method of classifying prognostic comorbidity in longitudinal studies: Development and validation. J Chronic Dis 40:373–383
20. Refsum H, Fisherstrand T, Guttormsen AB, Ueland PM (1997) Assessment of homocyteine status. J Inherit Metab Dis 20:286–294
21. Still RA, McDowell IF (1998) Clinical implications of plasma homocysteine measurement in cardiovascular disease. J Clin Pathol 51:183–188
22. Sahn DJ, DeMaria A, Kisslo J, Weyman A (1978) The committee on M-mode standardization of the American society of echocardiography. Recommendations regarding quantitation in M-mode echocardiography: results of a survey of echocardiographic measurements. Circulation 58:1072–1083
23. Bostom AG, Culleton BF (1999) Hyperhomocysteinemia in chronic renal disease. J Am Soc Nephrol 10:891–900
24. Chrysohoou C, Panagiotakos DB, Pitsavos C et al (2004) The associations between smoking, physical activity, dietary habits and plasma homocysteine levels in cardiovascular disease free people: the 'ATTICA' study. Vasc Med 9:117–123
25. Sobczak A, Wardas W, Zielinska-Danch W, Pawlicki K (2004) The influence of smoking on plasma homocysteine and cysteine levels in passive and active smokers. Clin Chem Lab Med 42:408–414
26. Tayama J, Munakata M, Yoshinaga K, Toyota T (2006) Higher plasma homocysteine concentration ia associated with more advanced ischemic arterial stiffness and greater blood pressure response to stress in hypertensive patients. Hypertens Res 29:403–409
27. Coban E, Ozdogan M, ErmiS C (2004) Plasma levels of homocysteine in patients with white-coat hypertension. Int J Clin Pract 58:997–999
28. Stehouwer CD, van Guldener C (2003) Does homocysteine cause hypertension? Clin Chem Lab Med 41:1408–1411
29. Aso Y, Yoshida N, Okumura K et al (2004) Coagulation and inflammation in overt diabetic nephropathy: association with hyperhomocysteinemia. Clin Chim Acta 348:139–145
30. Lentz SR (2005) Mechanisms of homcysteine induced atherothrombosis. J Thromb Haemost 3:1646–1654
31. Peeters AC, van Aken BE, Blom HJ, Reitsma PH (2007) den Heijer M (2007) The effect of homocysteine reduction

by B-vitamin supplementation on inflammatory markers. Clin Chem Lab Med 45:47–53
32. Schernthaner GH, Plank C, Minar E, Bieglmayer C, Koppensteiner R (2006) Schernthaner G (2006) No effect of homocysteine lowering therapy on vascular inflammation and haemostasis in peripheral arterial occlusive disease. Eur J Clin Invest 36:333–339
33. McGregor D, Shand B, Lynn K (2000) A controlled trial of the effect of folate supplements on homocysteine, lipids and hemorheology in end-stage renal disease. Nephron 85:215–220
34. Alvares Delfino VD, de Andrade Vianna AC, Mocelin AJ, Barbosa DS, Mise RA, Matsuo T (2007) Folic acid therapy reduces plasma homocysteine levels and improves plasma antioxidant capacity in hemodialysis patients. Nutiriton 23:242–247
35. Hirose S, Kono R, Mitarai T, Isoda K, Kim S, Shimoyama H (1999) Hyper-homocysteinemia in CAPD patients: peritoneal transport of total homocysteine at peritoneal equilibration test and daily elimination of total homocysteine. Nippon Jinzo Gakkai Shi 41:778–786
36. Vychytil A, Fodinger M, Papagiannopoulos M, Wölfl G, Hörl WH, Sunder-Plassmann G (1999) Peritoneal elimination of homocysteine moieties in continuous ambulatory peritoneal dialysis patients. Kidney Int 55:2054–2061
37. Churchill DN, Taylor DW, Keshaviah PR, The Canada-USA (CANUSA) Peritoneal Dialysis Study Group (1996) Adequacy of dialysis and nutrition in continuous ambulatory peritoneal dialysis patients. J Am Soc Nephrol 7:198–207
38. Johnson DW, Kay TD, Vesey DA, Isbel N, Campbell SB, Hawley CM (2000) Peritoneal homocysteine clearance is inefficient in peritoneal dialysis patients. Perit Dial Int 20:766–771
39. Loscalzo J (1996) The oxidant stress of hyperhomocysteinemia. J Clin Invest 98:5–7
40. Sydow K, Hornig B, Arakawa N et al (2004) Endothelial dysfunction in patients with peripheral arterial disease and chronic hyperhomocysteinemia: potential role of ADMA. Vasc Med 9:93–101
41. Cesari M, Zanchetta M, Burlina A et al (2005) Hyperhomocysteinemia is inversely related with left ventricular ejection fraction and predicts cardiovascular mortality in high risk coronary artery disease hypertensives. Arterioscler Thromb Vasc Biol 25:115–121
42. Laurent S, Boutouyrie P, Asmar R et al (2001) Aortic stiffness is an independent predictor of all-cause and cardiovascular mortality in hypertensive patients. Hypertension 37:1236–1241
43. Blacher J, Demuth K, Guerin AP, Safar ME, Moatti N, London GM (1998) Influence of biochemical alterations on arterial stifness in patients with end-stage renal disease. Arterioscler Thromb Vasc Biol 18:535–541
44. Zoungas S, Branley P, Kerr PG et al (2004) Aterosclerosis and folic acid supplementation trial in chronic renal failure: baseline results. Nephrology (Carlton) 9:130–141
45. Bortolotto LA, Safar ME, Billaud E et al (1999) Plasma homocysteine, aortic stiffness, and renal function in hypertensive patients. Hypertension 34:837–842
46. Woodside JV, McMahon R, Gallagher AM et al (2004) Total homocysteine is not a determinant of arterial pulse wave velocity in young healthy adults. Atherosclerosis 177:337–344