Inactive Matrix Gla Protein, Arterial Stiffness, and Endothelial Function in African American Hemodialysis Patients

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BACKGROUND

Matrix Gla protein (MGP) is a vascular calcification inhibitor dependent upon vitamin K for activation. Evidence suggests that elevated plasma inactive MGP levels (desphospho-uncarboxylated MGP, dp-ucMGP; indicating poorer vascular vitamin K status) are associated with greater cardiovascular disease (CVD) risk. Despite African Americans experiencing highest rates of kidney failure and CVD events, relationships between dp-ucMGP and CVD risk markers have not been examined in this population. We investigated vascular vitamin K status (*via* plasma dp-ucMGP) between African American hemodialysis (HD) patients and healthy controls, and the associations of dp-ucMGP with arterial stiffness and endothelial function in HD patients only.

METHODS

In 37 African American HD patients and 37 age- and race-matched controls, plasma dp-ucMGP was measured by enzyme immunoassay as a marker of vascular vitamin K status. Carotid-femoral pulse wave velocity (PWV; arterial stiffness measurement) and brachial artery flow-mediated dilation (FMD; endothelial function measurement) were assessed by applanation tonometry and ultrasound, respectively, in HD patients only.

RESULTS

Mean dp-ucMGP levels were 5.6 times higher in HD patients vs. controls (2,139 ± 1,102 vs. 382 ± 181 pmol/l, $P < 0.01$). Multiple linear regression, adjusting for age, sex, dialysis vintage, diabetes mellitus, CVD history, body mass index, and blood pressure, revealed that dp-ucMGP was independently related to PWV (standardized $\beta = 0.49$) and FMD (standardized $\beta = -0.53$) (both $P < 0.01$).

CONCLUSIONS

Our data suggest that the higher plasma dp-ucMGP concentrations found in African American HD patients may be associated with greater arterial stiffness and endothelial dysfunction.

Keywords: African American; arterial stiffness; blood pressure; chronic kidney disease; endothelial function; hypertension; vitamin K.

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The life span of adults with end-stage renal disease (ESRD) is reduced, and cardiovascular disease (CVD) accounts for approximately half the deaths among those undergoing hemodialysis (HD).[1] Vascular calcification is a key process in the development of atherosclerotic and arteriosclerotic CVD and contributes significantly to the greater mortality rates and CVD events in HD patients.[2] Mechanisms of vascular calcification in this population are not fully explained by traditional risk factors.[3] Therefore, identifying new biomarkers of this process would expand diagnostic utility and help improve clinical management in HD patients.

Recently, there has been interest in the vitamin K-dependent matrix Gla protein (MGP) and its putative role in regulating vascular calcification.[4] Vitamin K is necessary for the carboxylation of inactive MGP into active MGP, which in turn promotes binding to calcium ions.[5] It is postulated that carboxylated MGP counteracts vascular calcification in arteries and protects blood vessels from calcium overload.[6] In cases of vitamin K deficiency, when MGP is not activated, uncarboxylated MGP predominantly accumulates in areas of vascular calcification, and is associated with both intimal and medial calcification.[7] It has been suggested that desphospho-uncarboxylated MGP (dp-ucMGP) may be a suitable biomarker for *de novo* synthesis of uncarboxylated inactive MGP reflecting vascular vitamin K status, and may therefore contribute to CVD risk assessment.[4] In the chronic kidney disease setting, plasma dp-ucMGP levels have been reported to increase progressively from stage 2 onward, with the highest levels found in stage 5 patients on HD.[4,8] It is possible that higher plasma dp-ucMGP levels in HD patients reflect vascular vitamin K insufficiency such that the calcification-inhibitory activity of MGP is impaired, which may

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contribute to the higher CVD risk in this population. This notion is supported by few studies in HD patients, which have revealed that higher dp-ucMGP levels are associated with vascular calcification, CVD events, and mortality.[4,8–12]

As kidney failure rates are highest in African Americans, a limitation of the aforementioned investigations is that HD patients were predominately White. The US Renal Data System 2012 Report estimated that ESRD prevalence among African Americans was almost triple the rate for Whites.[13] Furthermore, rates of ESRD patients receiving HD were also considerably higher for African Americans (4,109 per million) than for Whites (757 per million).[13] Given that African Americans are particularly affected by CVD,[14] it is important to determine the association of plasma dp-ucMGP with vascular health in the African American HD patient population. Therefore, we determined vascular vitamin K status (*via* plasma dp-ucMGP) between African American HD patients and healthy controls, and the relations between dp-ucMGP and measures of arterial stiffness and endothelial function in HD patients only.

METHODS

Study participants

Participants in the cross-sectional investigation were 37 clinically stable African American ESRD patients (≥18 years of age) undergoing chronic HD who were recruited from Medical College of Georgia's outpatient dialysis (Augusta University, Augusta, Georgia). Exclusion criteria were: (i) had vascular procedures performed on both arms; (ii) had a medical condition (e.g., myocardial infarction or stroke in past 6 months, systemic cardiac failure, impaired autonomic control of blood pressure, or liver disease) or taking medications (e.g., anticoagulants, phosphodiesterase-5 inhibitors, β_1-selective β-blockers, nitrites, α-1 adrenergic blockers, erythromycin, and protease inhibitors) that could interfere with study results; (iii) women who were pregnant or breastfeeding; and (iv) unwilling or unable to provide informed consent. Data were obtained on demographic and clinical characteristics during the interview and from review of patient's files. The following measures were collected: age, sex, body mass index (BMI), dialysis vintage, etiology of ESRD, medical history of hypertension, diabetes mellitus and previous CVD, and smoking habit. Hypertension was defined as systolic blood pressure ≥140 mm Hg and/or diastolic blood pressure ≥90 mm Hg and/or use of antihypertensive medication. Diabetes mellitus status was obtained from electronic medical files and/or defined according to being in receipt of treatment for diabetes. CVD was defined as history of myocardial infarction, percutaneous coronary artery intervention, cardiac surgery, peripheral artery disease, or cerebrovascular disease. Smoking habit was defined if a patient currently smoked. In addition, 37 apparently healthy age- and race-matched controls were recruited from the Augusta, Georgia area to serve as a reference population. In this control group, plasma dp-ucMGP was measured. Study protocols were approved by the Institutional Review Board at Augusta University, and each participant provided written informed consent.

Biochemical measures

All HD patients were receiving dialysis 3 times per week for 3.5 to 5 hours. Venous blood samples were obtained before the start of a midweek HD session and after an overnight fast. Serum and citrated plasma were prepared after standard centrifugation and stored in a −80°C freezer until required for analysis. Serum concentrations of calcium, phosphorus, calcium-phosphorus product, albumin, hemoglobin, and intact parathyroid hormone were assessed by standard laboratory methods at Augusta University Medical Center. Dp-ucMGP was assessed in EDTA plasma using a previously described dual-antibody sandwich ELISA (VitaK, Maastricht University, The Netherlands).[4] The mean intra-assay and interassay coefficients of variation for dp-ucMGP were 5.6% and 9.9%, respectively.

Arterial stiffness

Carotid-femoral pulse wave velocity (PWV), an arterial stiffness measurement, was determined using the SphygmoCor system (AtCor Medical, Sydney, Australia) by sequentially recording electrocardiographic-gated carotid and femoral artery waveforms by applanation tonometry (Millar Instruments, Houston, TX). Using a segmometer, straight-line distance measurements were taken from the suprasternal notch to the carotid sampling site and from the suprasternal notch to the site where the femoral artery was measured. The time interval between the onset of femoral and carotid waveforms was determined as the mean from 10 consecutive cardiac cycles. The carotid-femoral PWV was calculated from the distance between measurement points (D, meters) and the measured time delay (t, seconds) between the peak of the ECG P-wave and the trough of a waveform as follows: carotid-femoral PWV = D/t (m/s).

Endothelial function

Endothelial function was determined by assessment of brachial artery flow-mediated dilation (FMD) following an overnight fast. Measurements took place in a quiet, temperature-controlled room (22–24°C), where patients were instructed to lie supine with their arm laterally extended for 20 minutes to establish a hemodynamic steady state. The brachial artery was imaged in duplex mode (simultaneous B-mode and blood velocity profiles) by a Doppler ultrasound (Hewlett-Packard/Phillips Sonos 5500, Andover, MA) using a 7.5-MHz linear array transducer placed 2 cm to 10 cm above the antecubital fossa. Blood velocity was obtained with the sample volume set at a depth between 1 cm and 3 cm. The average diameter and blood velocity for 30 cardiac cycles were recorded and analyzed to represent baseline values. Subsequently, a forearm occlusion cuff (D.E. Hokanson, Bellevue, WA) was placed immediately distal to the medial epicondyle and rapidly inflated to 240 mm Hg for 5 minutes (E-20 rapid cuff inflator; D.E. Hokanson) to induce arterial occlusion and reactive hyperemia of the brachial artery. ECG gaiting (Accusync 72; Accusync Medical Research, Milford, CT) was used to capture end-diastolic

arterial diameters for automated offline analysis of brachial artery vasodilatation (Brachial Analyzer Software, Medical Imaging Applications, Coralville, IA). FMD is expressed as a percent increase in peak diameter from baseline diameter.

Statistical analyses

Descriptive statistics are presented as mean ± SD for continuous variables and as frequency (percentage) for categorical variables. Normal distribution and homogeneity of variances were confirmed by Shapiro–Wilks *W* and Levene's tests, respectively. Differences in age, BMI, blood pressure, and dp-ucMGP levels between the HD patients and controls were tested using independent-samples *t*-test. Differences in proportions were tested using χ^2 test. For illustrative purposes, the HD patient group was subdivided into tertile groups of dp-ucMGP levels to visualize associations with dp-ucMGP. Linear trends across tertile groups were tested by analysis of variance with polynomial contrast for continuous variables and by Mantel–Haenszel linear-by-linear association χ^2 tests for categorical variables.

Bivariate correlations were used to examine associations of dp-ucMGP with carotid-femoral PWV and brachial artery FMD in the HD patients. We subsequently performed multiple linear regression analyses to determine whether dp-ucMGP was independently related to carotid-femoral PWV and brachial artery FMD after adjusting for potential confounding variables including age, sex, BMI, systolic blood pressure, dialysis vintage, diabetes mellitus, and history of CVD.[15,16] All analyses were conducted with SPSS software (version 24; IBM SPSS Statistics, Chicago, IL), and statistical significance was set at *P* value <0.05.

RESULTS

Plasma dp-ucMGP levels in HD patients and healthy controls

Age, sex, BMI, and diastolic blood pressure did not differ between the HD patients and controls (Table 1). However, the HD group vs. control group had higher levels of systolic blood pressure and plasma dp-ucMGP (both $P < 0.01$).

Table 1. Characteristics of the African American hemodialysis patients and healthy controls

Characteristic	HD patients	Controls	*P*
n	37	37	
Age, year	47.7 ± 10.4	47.7 ± 7.4	0.91
Male sex	32 (86.5)	29 (78.4)	0.83
Body mass index, kg/m^2	27.6 ± 6.9	25.8 ± 5.4	0.32
Systolic BP, mm Hg	144 ± 23	129 ± 14	<0.01
Diastolic BP, mm Hg	83 ± 14	82 ± 11	0.82
Plasma dp-ucMGP, pmol/l	2,139 ± 1,102	382 ± 181	<0.01

Values for categorical variables are given as number (percentage); values for continuous variables as means ± SD. Abbreviations: BP, blood pressure; dp-ucMGP, desphospho-uncarboxylated matrix Gla protein.

Characteristics of the HD patients and by tertiles of plasma dp-ucMGP

Descriptive characteristics of the HD patients are presented in Table 2. There were 37 African American HD patients, consisting of 32 males and 5 females with a mean age of 47.7 ± 10.4 years (range 19 to 78 years). The median dialysis vintage was 3.0 years (range 0.3 to 21.2 years). Of the 37 patients, the most common causes of ESRD were hypertension ($n = 20$), diabetes ($n = 6$), focal segmental glomerulosclerosis ($n = 4$), and glomerulonephritis ($n = 3$). Hypertension, previous history of CVD, and diabetes mellitus were observed in 95%, 60%, and 27%, respectively, of the HD patients. Mean concentrations of calcium, calcium × phosphorus product, albumin, and hemoglobin were within the National Kidney Foundation's Kidney Disease Outcomes Quality Initiative dialysis target ranges.[17] However, HD patients, on average, had elevated serum concentrations for phosphorus and parathyroid hormone.

Characteristics of the HD patients according to tertiles of dp-ucMGP are described in Table 2. Across increasing tertiles of dp-ucMGP, more HD patients had diabetes mellitus and a previous history of CVD (both $P < 0.05$). Furthermore, there were significant linear upward trends for age and carotid-femoral PWV across tertiles of dp-ucMGP (both $P < 0.01$). Conversely, a significant linear downward trend across tertiles of dp-ucMGP was observed for brachial artery FMD ($P < 0.01$). There were no differences in sex distribution, dialysis duration, hypertension prevalence, antihypertensive medication usage, smoking habits, BMI, blood pressure, serum calcium, serum phosphorus, serum Ca × P product, serum albumin, serum hemoglobin, or serum parathyroid hormone across tertiles of dp-ucMGP (all $P > 0.05$).

Associations of dp-ucMGP with carotid-femoral PWV and brachial artery FMD

Pearson's bivariate analyses demonstrated that dp-ucMGP was positively correlated with carotid-femoral PWV ($r = 0.44$) and negatively correlated with brachial artery FMD ($r = -0.52$) (both $P \leq 0.01$; Figure 1). Multiple linear regression models (with covariates age, sex, dialysis vintage, diabetes mellitus, history of CVD, BMI, and systolic blood pressure) revealed that dp-ucMGP was independently related to carotid-femoral PWV (standardized $\beta = 0.49$) and brachial artery FMD (standardized $\beta = -0.53$) (both $P < 0.01$). Table 3 shows that dp-ucMGP (14.3%), age (8.8%), sex (9.0%), diabetes mellitus (12.3%), and systolic blood pressure (22.4%) explained 66.8% of the variance in carotid-femoral PWV, with no contribution by dialysis vintage, history of CVD, and BMI. Table 4 shows that dp-ucMGP (16.8%), age (10.4%), and sex (12.1%) explained 39.3% of the variance in brachial artery FMD, with no contribution by dialysis vintage, diabetes mellitus, history of CVD, BMI, and systolic blood pressure.

DISCUSSION

To our knowledge, this is the first study investigating relationships between plasma dp-ucMGP and markers of

Table 2. Characteristics of the African American hemodialysis patients by tertiles of plasma dp-ucMGP

Plasma dp-ucMGP limits, pmol/l	Total sample	Low	Medium	High	*P*
		<1,540	1,540–2,660	>2,660	
n	37	12	13	12	
Age, year	47.7 ± 10.4	41.3 ± 11.5	46.0 ± 14.5	55.9 ± 7.1	<0.01
Male sex	32 (86.5)	9 (75.0)	12 (92.3)	11 (91.7)	0.22
Dialysis vintage, year	3.0 (0.3–21.2)	5.6 (0.4–21.2)	1.90 (0.3–7.2)	3.2 (0.5–7.6)	0.61
Hypertension	35 (94.6)	11 (91.7)	12 (92.3)	12 (100.0)	0.32
Diabetes mellitus	10 (27.0)	1 (8.3)	4 (30.8)	5 (41.7)	0.04
History of cardiovascular disease	22 (59.5)	5 (41.7)	7 (53.8)	10 (83.3)	0.04
Antihypertensive medication	33 (89.2)	10 (83.3)	12 (92.3)	11 (91.7)	0.51
Smoking habit	12 (32.4)	2 (16.7)	5 (38.5)	5 (41.7)	0.24
Body mass index, kg/m^2	27.6 ± 8.0	25.7 ± 8.4	29.7 ± 9.2	27.1 ± 6.0	0.66
Systolic blood pressure, mm Hg	144 ± 23	144 ± 30	144 ± 21	145 ± 20	0.84
Diastolic blood pressure, mm Hg	83 ± 14	84 ± 19	82 ± 12	82 ± 11	0.63
Serum calcium, mg/dl	9.2 ± 0.6	9.2 ± 0.7	9.2 ± 0.7	9.4 ± 0.5	0.54
Serum phosphorus, mg/dl	5.8 ± 1.6	5.8 ± 1.5	5.9 ± 1.7	5.7 ± 1.8	0.91
Serum Ca × P, mg^2/dl^2	53.2 ± 15.6	52.7 ± 14.2	53.4 ± 16.1	53.5 ± 17.6	0.90
Serum albumin, g/dl	4.2 ± 0.7	4.2 ± 0.2	4.3 ± 1.1	4.1 ± 0.3	0.80
Serum hemoglobin, g/dl	11.6 ± 1.2	11.7 ± 0.9	11.1 ± 1.1	12.2 ± 1.3	0.22
Serum PTH, pg/ml	409 ± 263	427 ± 292	444 ± 328	355 ± 136	0.53
Plasma dp-ucMGP, pmol/l	2,139 ± 1,102	1,003 ± 351	1,953 ± 256	3,445 ± 763	<0.01
Carotid-femoral PWV, m/s	9.3 ± 2.7	7.9 ± 1.6	8.9 ± 2.3	11.2 ± 3.2	<0.01
Brachial artery FMD, %	6.3 ± 4.3	9.3 ± 4.8	5.6 ± 3.4	4.0 ± 2.6	<0.01

Values for categorical variables are given as number (percentage); values for continuous variables, as means ± SD or median (range). Abbreviations: Ca × P, calcium-phosphorus product; dp-ucMGP, desphospho-uncarboxylated matrix Gla protein; FMD, flow-mediated dilation; PTH, parathyroid hormone; PWV, pulse wave velocity.

vascular health in African American HD patients, a population at greatest risk for CVD and mortality attributed in part to kidney failure. We found that dp-ucMGP levels were 5.6 times higher in African American HD patients compared to healthy controls. In the African American HD patients, dp-ucMGP levels were positively associated with arterial stiffness and negatively related to endothelial function. These relations were independent of potentially confounding factors such as age, sex, BMI, blood pressure, dialysis vintage, diabetes status, and CVD history. Collectively, our data suggest that the higher dp-ucMGP levels observed in the African American HD patients could be contributing to vascular dysfunction.

Several clinical reports have shown that plasma dp-ucMGP parallel the progression of CKD,[8,18] with the highest concentrations in HD patients.[4,9,10] Our results confirmed that dp-ucMGP concentrations are higher in HD patients compared to healthy controls, though this is the first time shown African American HD patients. The mean dp-ucMGP concentration in our African American HD population (2,139 ± 1,102 pmol/l) is comparable to the concentration observed by Cranenburg *et al.*[4] in 45 HD patients (2,126 ± 916 pmol/l), but lower than concentrations reported by Schlieper *et al.*[9] in 188 HD patients (2,850 ± 1,768 pmol/l) and Delanaye *et al.*[10] in 160 HD patients (2,704 ± 1,798 pmol/l). The relatively lower concentrations we observed may be attributed, in part, to our sampling of African American HD patients. Wei *et al.*,[18] in a recent population-based study, reported that dp-ucMGP levels were twice as low in Black vs. White South Africans. Similar racial differences in dp-ucMGP concentrations were also revealed in type 2 diabetes patients.[19] The mechanism by which race may affect the biologic activity of MGP is unknown. However, it is postulated that the Black population compared to other racial groups may have a reduced expression of MGP, which leads to lower levels of circulating dp-ucMGP.[19] Since our study did not include other racial groups, further investigation is needed to understand race as a factor on MGP synthesis and processing.

The most extensive vascular calcifications occur in CKD patients, especially those receiving HD.[2] Impaired inhibition of vascular calcification might be a major player underlying the high risk of CVD events in HD patients. Because MGP is a potent vascular calcification inhibitor, it has been suggested that dp-ucMGP may be useful in monitoring or even detecting vascular calcification in CKD and HD patients.[4,8] Indeed, positive

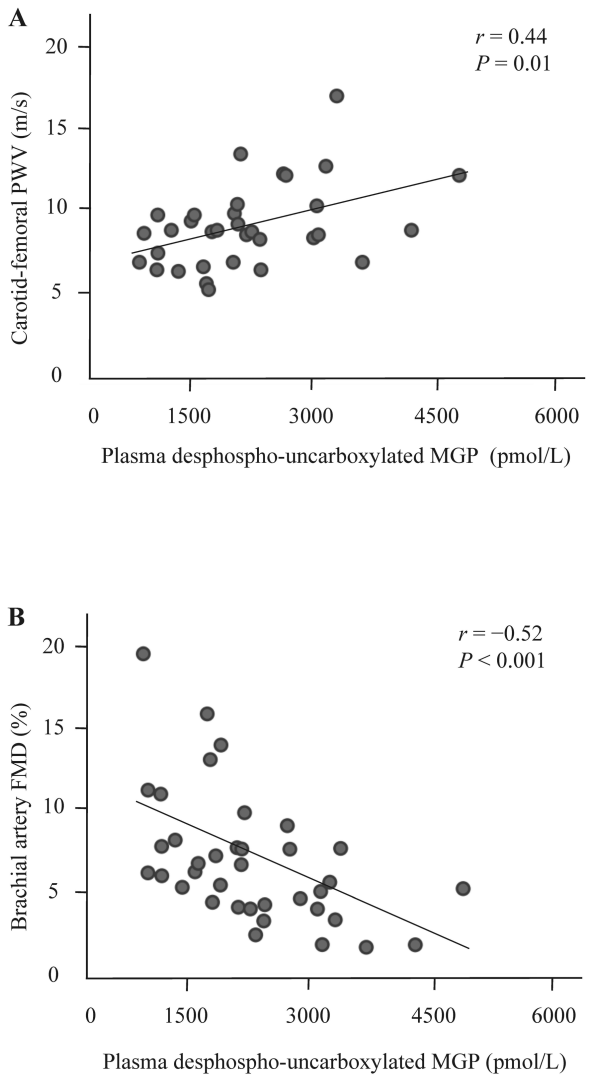

Figure 1. Bivariate correlations of plasma desphospho-uncarboxylated MGP concentrations with (**a**) carotid-femoral PWV and (**b**) brachial artery FMD in African American hemodialysis patients. Abbreviations: FMD, flow-mediated dilation; MGP, matrix Gla protein; PWV, pulse wave velocity.

correlations between plasma dp-ucMGP and vascular calcification have been reported in HD patients[10] and CKD patients not receiving dialysis.[8] Although vascular calcification was not assessed in our study, we found a positive relationship between circulating dp-ucMGP and arterial stiffness measured by PWV. A potential mechanism by which MGP could influence arterial stiffness is *via* vascular calcification.[20] Evidence suggests that vascular calcification is responsible for arterial stiffness and an increase in PWV.[21–24] The relationship between dp-ucMGP and PWV found in our HD patients reflects the findings of 2 recent population-based studies in Europe.[25,26] However, in 97 ESRD patients selected for kidney transplantation,[27] dp-ucMGP was not associated with PWV. This discrepancy could be because of their sample of patients eligible for transplantation, who are typically healthier than other ESRD patients, had lower

Table 3. Multiple linear regression model for dependent variable carotid-femoral PWV in African American hemodialysis patients

Independent variable	b ± SE	R^2
Intercept	−5.40 ± 2.96	
Age	0.088 ± 0.033[a]	0.088
Sex[b]	2.23 ± 0.78[a]	0.090
Dialysis vintage	NS	
Diabetes[c]	2.29 ± 0.66[a]	0.123
History of cardiovascular disease	NS	
Body mass index	NS	
Systolic blood pressure	0.065 ± 0.014[a]	0.224
Plasma dp-ucMGP	0.0012 ± 0.0003[a]	0.143
Total R^2		0.668

Abbreviations: b, multiple regression unstandardized coefficient; dp-ucMGP, desphospho-uncarboxylated matrix Gla protein; NS, not significant; R^2, proportion of variability in carotid-femoral pulse wave velocity (PWV) that is attributable to the regression equation. $n = 37$.
[a]$P < 0.05$.
[b]Sex coded such that males = 0 and females = 1.
[c]Diabetes coded such that no = 0 and yes = 1.

Table 4. Multiple linear regression model for dependent variable brachial artery FMD in African American hemodialysis patients

Independent variable	b ± SE	R^2
Intercept	2.25 ± 6.31	
Age	0.164 ± 0.071[a]	0.104
Sex[b]	4.16 ± 1.65[a]	0.121
Dialysis vintage	NS	
Diabetes	NS	
History of cardiovascular disease	NS	
Body mass index	NS	
Systolic blood pressure	NS	
Plasma dp-ucMGP	−0.002 ± 0.001[a]	0.168
Total R^2		0.393

Abbreviations: b, multiple regression unstandardized coefficient; dp-ucMGP, desphospho-uncarboxylated matrix Gla protein; NS, not significant; R^2, proportion of variability in brachial artery flow-mediated dilation (FMD) that is attributable to the regression equation. $n = 37$.
[a]$P < 0.05$.
[b]Sex coded such that males = 0 and females = 1.

scores and a smaller range of scores for arterial stiffness than other studies in HD patients, which may have limited the power of analysis.[27] Others have related arterial stiffness to total ucMGP levels in HD patients without assessing MGP phosphorylation. Such studies have had mixed results, either finding no association, or finding an inverse relation between total ucMGP and PWV.[27–30] It has been suggested that nonphosphorylated MGP is released into the circulation more easily than phosphorylated MGP.[4] Consequently, dp-ucMGP may be better

suited to assess vascular vitamin K status, and therefore contribute to CVD risk assessment.[4]

To the best of our knowledge, the relationship between endothelial function and plasma dp-ucMGP has not previously been investigated. Impaired endothelial function, reflected by lower FMD, has been linked to vascular calcification in both healthy populations and CKD patients.[31–33] FMD has also been reported to be lower in HD patients compared to healthy controls.[34,35] The negative relationship between plasma dp-ucMGP and FMD in our study suggests that endothelial function may be another vascular health marker potentially compromised due to the link between MGP and vascular calcification.

Strengths of this study were the robust assessments of arterial stiffness and endothelial function and the consideration of potential confounding variables. However, we acknowledge several study limitations. First, our small sample size limits the generalizability of our findings. Second, because of our cross-sectional study design, the associations between dp-ucMGP and vascular health measurements do not prove causality. In fact, it is plausible that elevated MGP is a consequence of CVD rather than causal, since vascular calcification and cardiac overload can promote MGP expression.[5–7] Also, the amount of dp-ucMGP in circulation depends on the total amount of MGP available.[4] Given that HD patients are likely to produce more MGP,[4,12] the associations of dp-ucMGP with PWV and FMD may be related to overall MGP status rather than vitamin K status. Only a vitamin K intervention trial in HD patients could elucidate the role of MGP carboxylation on markers of vascular health. To date in HD patients, 2 vitamin K supplementation trials have demonstrated dose-dependent decreases in inactive MGP levels,[36,37] but whether vitamin K-dependent MGP activation translates into relevant CVD-related outcomes remains to be determined. Notwithstanding, evidence from a vitamin K trial in non-HD patients suggest that dp-ucMGP in circulation is not related to coronary artery calcification progression, even though vitamin K supplementation reduced circulating dp-ucMGP and the progression of coronary artery calcification.[38,39] Another limitation is that we did not assess additional markers of vitamin K status (e.g., phylloquinone and PIVKA-II), which would have provided support that high inactive MGP levels reflect subclinical vitamin K insufficiency. Dp-ucMGP has been proposed as a vascular vitamin K status marker; however, evidence is currently insufficient to support what levels of dp-ucMGP are required for optimal functioning, and thus more research is needed. Lastly, we did not assess vitamin K intake, which would have provided information on whether inadequate vitamin K intake leads to poor vascular health through an increase in plasma dp-ucMGP. However, because of its lipophilic properties and incorporation into lipoproteins, vitamin K is not expected to be removed by HD treatment.[40] Therefore, it is plausible that the higher dp-ucMGP levels in our HD patients is due to inadequate vitamin K intake.

In conclusion, our data suggest that the higher plasma dp-ucMGP concentrations found in African American HD patients may be associated with greater arterial stiffness and endothelial dysfunction. Whether our study findings reflect suboptimal vitamin K status or increased MGP synthesis is not known.

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DISCLOSURE

M.H.J.K. and C.V. are employees of the R&D Group VitaK, Maastricht University. Other authors declared no conflict of interest.

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