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Vascular endothelial growth factor, its soluble receptor, and hepatocyte growth factor: clinical and genetic correlates and association with vascular function

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Aims Growth factors play an important role in regulating vascular function. Data are limited regarding clinical and genetic correlates of endothelial growth factors and their associations with vascular function.

Methods and results We evaluated clinical and genetic correlates of circulating vascular endothelial growth factor A (VEGF), its soluble receptor sFlt-1, and hepatocyte growth factor (HGF) in 3754 Framingham Study participants. We also related the growth factors to measures of brachial artery function. Serum VEGF and HGF were higher and sFLt-1 was lower in women and smokers. VEGF and HGF were associated positively with body mass index; both displayed strong positive associations with the metabolic syndrome ($P < 0.001$) and its components. The heritabilities of VEGF, sFlt-1, and HGF were 78, 13, and 38%, respectively. VEGF and HGF were related positively to baseline brachial diameter ($P < 0.01$) and to baseline mean flow velocity ($P < 0.001$) in age- and sex-adjusted models, but the multivariable models failed to reach significance. None of the growth factors were related to flow-mediated dilation.

Conclusion In our community-based sample, circulating VEGF and HGF demonstrated high heritabilities and a sexual dimorphism. Increased angiogenesis and greater endothelial cell turnover may underlie associations of these growth factors with risk factors including smoking.

Keywords Vascular growth factors • VEGF • SFlt-1 • HGF • Vascular function • Heritability • Metabolic syndrome

Introduction

Cardiovascular disease (CVD) is a major cause of morbidity and mortality in the United States.[1] Substantial scientific evidence identified progressive structural and functional changes in the vessel wall—termed as vascular remodelling[2]—as hallmarks of atherosclerosis,[3] which precedes overt CVD by decades. As a consequence, investigating the determinants of vascular remodelling and identifying biomarkers of the process are critical.

Vascular endothelial growth factor (VEGF) modulates physiological and pathophysiological vascular development.[4] VEGF stimulates the production of nitric oxide (NO) and prostacyclin by endothelial cells,[5,6] increases vascular permeability,[7] stimulates growth,[8] and prevents apoptosis of endothelial cells. Elevated circulating VEGF has

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been observed in hypertension,[9] coronary disease,[10] myocardial infarction,[11] peripheral arterial disease,[10] and heart failure.[12] VEGF binds two receptors, VEGFR1 and VEGFR2.[13] The soluble isoform of VEGFR1 (sFlt-1) is found in circulation where it inhibits VEGF by direct sequestration.[13]

Hepatocyte growth factor (HGF) is another angiogenic growth factor expressed by multiple cell types, including endothelial and vascular smooth muscle cells.[14] Increased circulating HGF has been reported in hypertension,[15] obesity,[16] myocardial infarction,[17] ventricular hypertrophy,[18] and heart failure.[19] HGF is also positively associated with arterial stiffness and with reactive hyperaemia in hypertension,[20] and has been related to carotid artery remodelling.[21]

Although VEGF, sFlt-1, and HGF play a fundamental role in vascular remodelling, the clinical correlates of circulating VEGF, sFlt-1, and HGF have not been comprehensively assessed. We related VEGF, sFlt-1, and HGF levels to CVD risk factors and to measures of brachial artery (BA) function, and estimated the heritabilities of these biomarkers. We hypothesized that: a higher burden of CVD risk factors is associated with higher VEGF and HGF but lower sFlt-1; these biomarkers are heritable; VEGF and HGF are associated with better endothelial function but sFlt-1 is associated with endothelial dysfunction.

Methods

Study sample

The design of the Framingham Offspring Study has been described elsewhere.[22] Starting in 2002, 4095 participants with at least one parent in the Offspring cohort were enrolled in the Generation 3 cohort (Examination 1).[23] We excluded 341 attendees for reasons detailed in the Supplementary material online. After exclusions, 3754 participants remained eligible. At their first examination, Generation 3 participants have been comprehensively phenotyped as detailed in the Supplementary material online. All participants provided informed consent and the study complies with the Declaration of Helsinki and was approved by the Institutional Review Board at the Boston University Medical Center.

Laboratory measurements of vascular endothelial growth factor, sFlt-1, and hepatocyte growth factor

Blood was drawn after an overnight fast, immediately centrifuged and stored at -80°C until biomarkers were assayed. Serum VEGF, sFlt-1, and HGF were measured with commercial assays (R&D Inc.) as detailed in the Supplementary material online.

Flow-mediated dilation and reactive hyperaemia measurements

Baseline and hyperaemic measures of BA structure and function have been determined using a Toshiba SSH-140A ultrasound system as described previously[24] and detailed in the Supplementary material online.

Statistical analyses

Additional details on the analyses performed are displayed in the Supplementary material online.

Clinical correlates

VEGF, sFlt-1, and HGF were natural log-transformed to normalize their distributions. To assess the association of each biomarker with clinical covariates [age, sex, systolic and diastolic BP, anti-hypertensive medication, diabetes, total cholesterol, high-density lipoprotein (HDL) cholesterol, triglycerides, smoking, body mass index (BMI), alcohol consumption, estimated glomerular filtration rate (eGFR)], we first identified predictors for each biomarker separately using stepwise forward multivariable linear regression ($P \leq 0.1$ for model entry). The associations of each biomarker with variables that were significant in the stepwise forward regression analysis ($P < 0.05$) were then examined using the generalized estimating equations (GEE; using the Compound Symmetry Correlation Matrix) to account for relatedness among participants.

In secondary analyses, we related biomarkers to select risk factors that are categorical and frequently used in clinical practice: Hypertension, BP $\geq$140/90 mmHg or anti-hypertensive treatment; obesity, BMI $\geq$30 kg/m^2; abdominal obesity, waist circumference $\geq$102 cm (men) or $\geq$89 cm (women); low HDL cholesterol, $<$40 mg/dL (men) or $<$50 mg/dL (women); high triglycerides, $\geq$150 mg/dL or lipid-lowering treatment. Age and eGFR were modelled as continuous variables, all other significant continuous traits were replaced by their binary counterpart in secondary analyses. We also related VEGF and HGF to prevalence of the metabolic syndrome (MetS), adjusting for age, sex, and smoking. MetS was defined by the presence of $\geq$3 of: increased waist circumference; elevated BP [$\geq$130 (systolic) or $\geq$85 mmHg (diastolic), or anti-hypertensive treatment]; hyperglycaemia (fasting glucose $\geq$100 mg/dL or treatment for elevated glucose); hypertriglyceridaemia ($\geq$150 mg/dL or lipid-lowering treatment); low HDL cholesterol [$<$40 (men), $<$50 mg/dL (women)].[25] A two-sided *P*-value below 0.05 was considered statistically significant.

Heritability estimates

As detailed in the Supplementary material online, heritability for each log-biomarker was estimated using variance components analysis. The analyses were performed on residuals from models adjusting for (1) age and sex; (2) age, sex, and all other covariates that were significantly associated with the respective biomarker in our prior analyses (clinical correlates).

Relations to vascular function

Log-biomarker concentrations were related to four vascular function measures using GEE: baseline BA diameter, baseline BA mean flow velocity, flow-mediated dilation (FMD), and hyperaemic mean flow velocity. Model 1 adjusted for age and sex; model 2 adjusted for age, sex, mean arterial pressure, pulse pressure, heart rate, BMI, total/HDL cholesterol, fasting glucose, diabetes, smoking within 6 h prior to the procedure, prevalent CVD, hormone replacement therapy, hypertension, lipid-lowering medication, and walk test. These covariates correlate with BA vascular function in our cohort.[24]

Results

Information on the number of siblings per family in the study sample is provided in Supplementary material online, *Table S1*. The characteristics of our study sample are shown in *Table 1*. After adjusting for age and sex, LogVEGF correlated weakly with LogsFlt-1 ($r = 0.024$) and moderately with LogHGF ($r = 0.18$). The age- and sex-adjusted correlation between LogsFlt-1 and LogHGF was low ($r = 0.14$).

Clinical correlates of vascular endothelial growth factor, sFlt-1, and hepatocyte growth factor

In multivariable analyses, VEGF had significant positive associations with female sex, systolic BP, smoking, and BMI (*Table 2*). Replacing all significantly associated continuous variables by their categorical counterparts, VEGF was positively associated with abdominal obesity ($P < 0.001$), hypertension ($P = 0.02$), and MetS

Table 1 Characteristics of the study sample

Variables	Women ($n = 2002$)	Men ($n = 1752$)
Clinical features		
Age, years	40 ± 9	40 ± 9
Systolic BP, mmHg	113 ± 14	121 ± 12
Diastolic BP, mmHg	73 ± 9	78 ± 9
BMI, kg/m^2	25.9 ± 6	27.8 ± 4.6
Waist circumference, inches	34.8 ± 6.1	38.6 ± 5
Smoking, %	14	16
Alcohol consumption, ounces per month	5.8 ± 7.9	13.6 ± 17.9
eGFR, mL/min/1.73 m^2	99 ± 19	100 ± 17
Hypertension, %	12	20
Diabetes, %	2	3
UACR, mg/g	9.2 ± 24.8	5.9 ± 12.6
Biochemical features		
Total cholesterol, mg/dL	185 ± 34	193 ± 37
HDL cholesterol, mg/dL	61 ± 16	47 ± 12
Triglycerides, mg/dL	97 ± 63	134 ± 105
VEGF, ng/mL	285 (159,471)	274 (157,437)
sFlt-1, ng/mL	136 (100,184)	149 (112,194)
HGF, ng/mL	823 (696,970)	814 (700,965)
Metabolic syndrome, %	14	26.6
Vascular function	$N = 1860$	$N = 1674$
Baseline brachial diameter, mm	3.50 ± 0.44	4.76 ± 0.61
FMD, %	7.12 ± 3.81	4.57 ± 2.94
Baseline mean flow velocity, cm/s	6.76 ± 3.58	7.95 ± 4.71
Hyperaemic mean flow velocity, cm/s	65.9 ± 18.0	57.7 ± 17.6

Values represent mean ± SD for continuous variables and % for discrete. Values for VEGF, sFlt-1, and HGF represent median (25th percentile, 75th percentile).
BMI, body mass index; BP, blood pressure; SD, standard deviation; eGFR, estimated glomerular filtration rate; HDL, high-density lipoprotein; HGF, hepatocyte growth factor; sFlt-1, soluble fms-like tyrosine kinase-1; UACR, urinary albumin to creatinine ratio; FMD, flow-mediated dilation; VEGF, vascular endothelial growth factor.

($P < 0.001$). Consistent with this finding, VEGF increased with the number of components of MetS (*Figure 1*). In secondary analyses, VEGF concentrations did not differ between pre- and post-menopausal women, nor between women in the follicular vs. luteal phases of their menstrual cycle (data not shown). Serum sFlt-1 levels were positively associated with male sex and age and negatively associated with smoking and renal function (*Table 2*).

HGF displayed positive associations with age, female sex, diastolic BP, anti-hypertensive treatment, diabetes, triglycerides, smoking, and BMI, and an inverse association with HDL cholesterol (*Table 2*). In secondary analyses, replacing significant continuous traits by binary variables, HGF was positively associated with hypertension ($P < 0.001$), diabetes ($P < 0.001$), high triglycerides ($P < 0.001$), smoking ($P < 0.001$), abdominal obesity ($P < 0.001$), and the MetS ($P < 0.001$). HGF levels increased gradually with the number of components of the MetS ($P < 0.001$; *Figure 1*).

Table 2 Clinical correlates of VEGF, sFlt-1, and HGF

	Estimate	95% CI	*P*-value
Dependent: VEGF			
Age, years	0.023	(−0.005,0.051)	0.10
Sex, m vs. f	−0.120	(−0.168,−0.073)	<0.001
Smoking, yes vs. no	0.127	(0.065,0.190)	<0.001
Systolic BP, mmHg	0.033	(0.007,0.060)	0.014
Triglycerides, mg/dL	0.026	(−0.0004,0.052)	0.054
BMI, kg/m^2	0.074	(0.048,0.100)	<0.001
Dependent: sFlt-1			
Age, years	0.022	(0.001,0.043)	0.041
Sex, m vs. f	0.110	(0.074,0.147)	<0.001
Smoking, yes vs. no	−0.103	(−0.161,−0.045)	<0.001
eGFR, mg/g	−0.027	(−0.049,−0.005)	0.017
Dependent: HGF			
Age, years	0.027	(0.018,0.036)	<0.001
Sex, m vs. f	−0.041	(−0.059,−0.023)	<0.001
Diastolic BP, mmHg	0.013	(0.004,0.022)	0.004
Anti-hypertensive medication, yes vs. no	0.044	(0.013,0.075)	0.005
Diabetes, yes vs. no	0.090	(0.039,0.140)	<0.001
High-density lipoprotein cholesterol, mg/dL	−0.01	(−0.019,−0.001)	0.039
Triglycerides, mg/dL	0.01	(0.001,0.019)	0.039
Smoking, yes vs. no	0.115	(0.092,0.139)	<0.001
BMI, kg/m^2	0.053	(0.044,0.063)	<0.001

Estimates reflect increase in log-biomarker level per one-SD increase of the continuous traits.
BMI, body mass index; BP, blood pressure; SD, standard deviation; SE, standard error; eGFR, estimated glomerular filtration rate; HGF, hepatocyte growth factor; sFlt-1, soluble fms-like tyrosine kinase-1; VEGF, vascular endothelial growth factor; CI, confidence interval of estimate.

Heritability estimates for vascular endothelial growth factor, sFlt-1, and hepatocyte growth factor

For VEGF, the estimated heritability ranged from 0.77 to 0.78. For sFlt-1, the heritabilities were lower at 0.12–0.13. The heritability for HGF was 0.37–0.38 (*Table 3*).

Association of vascular endothelial growth factor, sFlt-1, and hepatocyte growth factor with vascular function

In age- and sex-adjusted models, VEGF and HGF were associated positively with baseline BA diameter and with baseline BA mean flow velocity. These associations were no longer significant after adjusting for covariates (*Table 4*). Adding BMI alone or heart rate and BMI to the models was sufficient to render statistically non-significant the associations of growth factors with vascular function (noted in age- and sex-adjusted models) in most instances. sFlt-1 was not associated with any measure of vascular structure or function (*Table 4*).

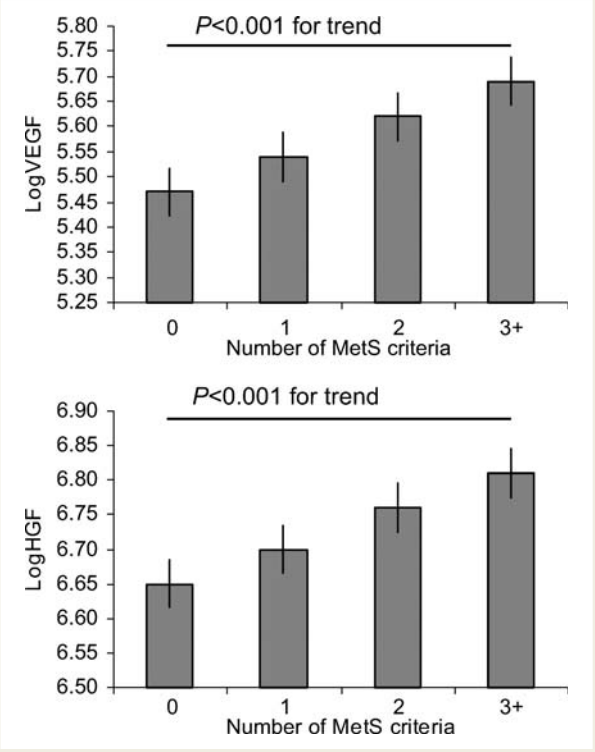

Figure 1 Multivariable adjusted means for log-transformed vascular endothelial growth factor (logVEGF) and hepatocyte growth factor (logHGF) stratified by the number of criteria for the metabolic syndrome (MetS).

Table 3 Heritability estimates for VEGF, sFlt-1, and HGF

Biomarker	Model	Heritability	95% CI	*P*-value
HGF	Age- and sex-adjusted	0.37	(0.29,0.45)	<0.001
	Multi-variable adjusted	0.38	(0.29,0.46)	<0.001
sFlt-1	Age- and sex-adjusted	0.12	(0.05,0.19)	<0.001
	Multi-variable adjusted	0.13	(0.05,0.20)	<0.001
VEGF	Age- and sex-adjusted	0.77	(0.71,0.84)	<0.001
	Multi-variable adjusted	0.78	(0.71,0.85)	<0.001

Multivariable models were adjusted for those variables, which were significant for the respective biomarkers in *Table 2*.
HGF, hepatocyte growth factor; sFlt-1, soluble fms-like tyrosine kinase-1; VEGF, vascular endothelial growth factor; CI, confidence interval.

Discussion

Principal findings

First, we observed a sexual dimorphism in circulating biomarkers: VEGF and HGF concentrations were higher in women, whereas sFlt-1 was lower. Second, both VEGF and HGF were highly significantly and positively associated with select risk factors, i.e. smoking, BMI, and obesity, and with the constellation identifying MetS. sFlt-1 was inversely related to smoking. Third, we observed very high heritability for VEGF, moderately high heritability for HGF, and modest heritability for sFlt-1. Finally, VEGF and HGF were associated with baseline brachial diameter and baseline mean flow velocity in age- and sex-adjusted models. However, adjustment for clinical covariates rendered these associations non-significant, suggesting that the associations of growth factors with baseline measures of vascular structure and function may be mediated via their relations to other risk factors.

Comparison with the literature

Sexual dimorphism of circulating vascular endothelial growth factor, hepatocyte growth factor, and sFlt-1

Sexual dimorphism for vascular growth factors has previously been reported in a relatively small case–control study of obese vs. non-obese individuals.[26] In that study, circulating VEGF-C, VEGF-D, and angiopoetin-2 were significantly higher in women.[26] Yamamoto *et al.*[21] observed no association of plasma HGF with sex. Recently, one study reported higher VEGF in men,[27] whereas another observed increased concentrations in women.[28] Our results are in agreement with the latter findings. We observed significantly higher VEGF and HGF and lower sFlt-1 in women.

The mechanisms for these sex-related differences in circulating growth factors are not clear, but experimental studies suggest that sex hormones may modify vascular growth factor levels. Endogenous estrogens have vasculo-protective effects, in part by regulating VEGF expression.[29] For example, estrogen modulates VEGF and sFlt-1 expression in the endometrium,[30] in cancer cells,[31] and in vascular smooth muscle cells *in vitro*.[32] Estrogens also increased HGF production.[33] It is conceivable, therefore, that higher endogenous estrogen contributes to increased VEGF and HGF in women.

Association of vascular endothelial growth factor, sFlt-1, and hepatocyte growth factor with cardiovascular disease risk factors

The strong positive associations of VEGF and HGF concentrations with obesity and presence of the MetS are consistent with prior reports.[16,34] Statistically significant correlations have been reported between VEGF or HGF and lipid levels/dyslipidaemia in some[27,34,35] but not in other studies.[21] Importantly, most prior studies had relatively small samples, limiting their statistical power to detect modest associations. In our large community-based sample, we noticed a borderline significant positive association of VEGF with triglycerides, a significant positive association of HGF with triglycerides, and a significant negative association of HGF with HDL cholesterol. We also observed strong associations between biomarkers and smoking. VEGF and HGF were higher in smokers, whereas sFlt-1 levels were lower. These results are in agreement with most[27,36] but not all previous studies.[37,38]

Table 4 Association of VEGF, sFlt-1, and HGF with measures of vascular function

	Beta	95% CI	*P*-value	Beta	95% CI	*P*-value
	Baseline brachial diameter			Flow-mediated dilation		
Age- and sex-adjusted						
VEGF	0.03	(0.01,0.05)	0.0049	−0.05	(−0.20,0.09)	0.47
HGF	0.11	(0.05,0.18)	0.001	−0.002	(−0.44,0.44)	0.99
sFlt-1	0.03	(−0.004,0.06)	0.09	0.09	(−0.11,0.29)	0.38
Multivariable-adjusted						
VEGF	0.01	(−0.01,0.03)	0.29	−0.05	(−0.19,0.10)	0.52
HGF	−0.02	(−0.09,0.04)	0.50	0.01	(−0.45,0.46)	0.98
sFlt1-1	0.02	(−0.005,0.05)	0.11	0.05	(−0.14,0.25)	0.59
	Baseline mean flow velocity			Hyperaemic mean flow velocity		
Age- and sex-adjusted						
VEGF	0.35	(0.17,0.53)	<0.001	0.07	(−0.68,0.83)	0.85
HGF	2.17	(1.64,2.70)	<0.001	0.31	(−1.98,2.59)	0.79
sFlt-1	−0.05	(−0.29,0.19)	0.68	−0.51	(−1.55,0.53)	0.33
Multivariable-adjusted						
VEGF	0.02	(−0.14,0.19)	0.81	−0.24	(−1.00,0.53)	0.55
HGF	0.07	(−0.45,0.59)	0.80	−1.84	(−4.27,0.58)	0.14
sFlt1-1	−0.09	(−0.31,0.13)	0.42	−0.64	(−1.67,0.40)	0.23

Beta: regression coefficient. Regression coefficient indicates increase of baseline brachial diameter in millimetres, of flow-mediated dilation in %, baseline and hyperaemic mean flow in cm/s per one-unit increase in log biomarker. For example, a 2.72-fold ($2.72 = e^1$) increase in VEGF is associated with an increase of 0.03 mm in baseline brachial diameter. HGF, hepatocyte growth factor; sFlt-1, soluble fms-like tyrosine kinase-1; VEGF, vascular endothelial growth factor; confidence interval.

Previous clinical studies reported higher growth factors in hypertensives.[9,15] In agreement with these results, we observed positive associations of VEGF and HGF concentrations with hypertension, and with BP variables modelled as continuous traits.

Finally, we observed a positive association of HGF and sFlt-1 levels with age. This is consistent with a prior report by Yamamoto *et al.*[21]

Although the different biomarkers were in part related to the same clinical correlates (e. g. BP and BMI), the correlation between the biomarkers themselves was rather low. This might be explained by the fact that traits like BP or BMI are influenced by multiple mechanisms.

Furthermore, the lack of correlation between serum levels of the three biomarkers is intriguing, but it does not negate potential correlations among the markers at the tissue level.

Possible mechanisms for association of vascular endothelial growth factor and hepatocyte growth factor with cardiovascular disease risk factors

Increasing evidence links vascular growth factors to CV risk factors, subclinical, and overt atherosclerotic disease. Clinical studies observed elevated circulating VEGF and HGF in patients with various forms of CVD.[10–12,17,19] Furthermore, growth factor levels have been associated with subclinical atherosclerotic disease.[18,21] It is controversial, however, whether vascular growth factors have a proatherogenic or a vasculoprotective net effect.[39]

HGF and VEGF and their receptors are found in atherosclerotic lesions,[40–42] and VEGF promoted plaque progression in animal models.[43] Other studies support a vasculoprotective role of VEGF[44] and VEGF gene transfer has been discussed as a new treatment option for ischaemic heart disease.[45]

Our data indicate that circulating growth factors are related to several CVD risk factors and thus underscore that these biomarkers might play an important role in atherosclerotic diseases, either as disease markers or as contributors to the disease process. In particular, we found a prominent positive association of circulating HGF and VEGF with obesity and the MetS. Production of both biomarkers within the adipose tissue[16,46] may explain this finding. An alternative possibility is that vascular growth factors may play a key role in the regulation of adipogenesis.[47]

We also observed a positive association of HGF and VEGF with hypertension. Impaired vasculogenesis has been proposed as a contributing factor to hypertension and VEGF application has been reported to induce hypotension in animal models[48] and in clinical trials.[49] Furthermore, hypertension is a known side-effect of VEGF-pathway inhibitors, e. g. in the setting of different cancer therapies.[50,51] Overall, these data are consistent with the notion that VEGF may be a marker for hypertension, with higher circulating levels representing a response to high systemic BP. The observed association between all three biomarkers and smoking may be because smoking promotes atherosclerosis/inflammation, also known to increase growth factor expression.[52] Of note, ageing is associated with modifications in the vessel wall, which are in many ways mechanistically comparable to

changes in atherosclerosis.[53] This might explain the associations of sFlt-1 and HGF levels with age.

Heritability of vascular endothelial growth factor, sFlt-1, and hepatocyte growth factor

To the best of our knowledge, this is the first study to report heritability estimates for sFlt-1 and HGF. The heritability for sFlt-1 was relatively low, that of HGF was moderate. In contrast, VEGF displayed substantial heritability. Consistent with our data, two previous studies reported very high heritability for VEGF.[54,55] Overall, these data are consistent with the notion that angiogenesis/collateral formation is a heritable phenotype.

Association of vascular endothelial growth factor, sFlt1, and hepatocyte growth factor with vascular function

Data on the association of vascular growth factors with vascular function are relatively limited. Flow-mediated dilation was impaired in a small series of hypertensive patients and correlated negatively with circulating VEGF.[56] Schmidt-Lucke *et al.*,[38] similarly found an inverse correlation between circulating VEGF and endothelial function.

To our knowledge, relations of circulating VEGF, sFlt-1, and HGF to indices of vascular function in the community have not been reported. We observed an association of VEGF and HGF with baseline measures of vascular structure (BA diameter) and function (BA mean flow velocity). Interestingly, these associations were attenuated upon adjustment for other clinical covariates, suggesting that the association between VEGF and HGF and vascular structure and function may be mediated via these risk factors.

Limitations

Limitations of our investigation include the cross-sectional design. Also, the sample was predominantly white, which limits the generalizability of our findings. Given that platelets are an important source of VEGF, it would be desirable to adjust levels for platelet counts. We were unable to do so because platelet counts were not measured in our samples. Each biomarker was measured only on a single occasion in each participant, which might have led to some random misclassification biasing our results towards the null hypothesis. Due to the exploratory character of the present analyses, we did not adjust for multiple testing and acknowledge that our findings require confirmation in other cohorts.

Conclusions

In our investigation of a large community-based sample, we observed that VEGF, its soluble receptor sFlt-1, and HGF are heritable. VEGF and HGF display associations with established vascular risk factors, including the cluster defining the MetS. Additional studies are warranted to confirm these findings.

Supplementary material

Supplementary material is available at *European Heart Journal* online.

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Conflict of interest: G.F.M. is owner of Cardiovascular Engineering Inc., a company that designs and manufactures devices that measure vascular stiffness.

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