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ORIGINAL ARTICLE

Hepatocyte growth factor and risk of incident stroke in Black and White Americans in the Reasons for Geographic and Racial Differences in Stroke study

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Abstract

Background: Hepatocyte growth factor (HGF) is a cytokine produced in response to endothelial damage. Higher levels correlate with cardiovascular risk factors, including hypertension and diabetes.

Objectives: We hypothesized that HGF is associated with stroke.

Methods: The Reasons for Geographic And Racial Differences in Stroke (REGARDS) study enrolled 30,239 Black and White Americans aged ≥45 years from 2003 to 2007. In this case-cohort study, after 5.5 years of follow-up, circulating baseline HGF was measured in 557 participants with incident ischemic stroke and in a cohort random sample of 964 participants. Hazard ratios (HRs) per SD log-transformed HGF and by HGF quintile were calculated using Cox proportional hazards models adjusting for stroke risk factors and other correlates of HGF. Differences by race and sex were tested using interaction terms.

Results: Median HGF was 295 (IQR, 209-402) pg/mL. HGF was higher with older age, male sex, prevalent cardiovascular disease, smoking, and warfarin use, but did not differ by race. The adjusted HR of incident ischemic stroke per SD higher baseline HGF (145 pg/mL) was 1.30 (CI, 1.00-1.70), with no difference by sex or race. HGF in the highest (>434 pg/mL) vs lowest quintile (<135 pg/mL) was associated with an adjusted HR of incident stroke of 2.12 (CI, 1.31-3.41).

Conclusion: In the REGARDS study, higher HGF was associated with increased risk of incident ischemic stroke in Black and White adults, with a doubling in risk of HGF in the top quintile compared with the lowest quintile after adjusting for other stroke risk factors.

KEYWORDS
biomarker, hepatocyte growth factor, ischemic stroke, longitudinal studies, risk factors

Essentials

- Hepatocyte growth factor (HGF) is produced in response to blood vessel damage.
- We studied whether higher baseline HGF is associated with incident stroke in a large cohort study.
- Higher HGF was strongly associated with stroke independent of other stroke risk factors.
- Participants in the highest quintile HGF had double risk of stroke compared with the lowest.

1 | INTRODUCTION

Stroke is a leading cause of morbidity and mortality in the United States, with Black Americans having increased risk (relative to White Americans), especially between ages 45 and 65 years [1]. Vascular remodeling and endothelial dysfunction may precede overt stroke for decades [2], so identifying biomarkers may provide the opportunity for early detection and prevention through development of new preventive interventions. Hepatocyte growth factor (HGF) is a mitogenic cytokine produced in response to tissue injury and endothelial dysfunction [3]. It is active in cell growth/embryonic development, motility, wound healing, and angiogenesis [4] and may play a role in response to atherosclerotic disease through proangiogenic, anti-inflammatory, and antifibrotic mechanisms [5]. HGF is a tyrosine kinase receptor agonist and signals through the MET pathway, making it a possible target for clinical therapeutics [6,7].

Plasma HGF is elevated in individuals with established cardiovascular and metabolic risk factors, including hypertension [8], obesity [9], diabetes [10], and left ventricular hypertrophy (LVH) [11], but whether it is a causal risk factor is unknown. It was previously studied as a marker of disease severity in heart failure [12,13] and stroke [14] (especially in those with dyslipidemia [15]) and is associated with progression of atherosclerosis [16]. HGF has also been studied as a possible contributor to racial disparities in cardiovascular disease. In the Multi-Ethnic Study of Atherosclerosis (MESA), investigators found higher HGF was associated with more coronary calcium, and this association was strongest in Black participants [17], suggesting that HGF may account for some racial disparities in cardiovascular disease. They also identified genetic differences in expression of HGF across race and ethnicity (26), which could potentially explain differences in cardiovascular disease.

Two prospective studies reported associations of higher HGF with incident stroke, the Women's Health Initiative (WHI) and MESA. WHI included only postmenopausal women [18], limiting its generalizability. In MESA, the association of HGF with stroke risk did not differ by race [19], but this important analysis was limited by low power.

In this study, to further elucidate mechanisms of stroke, we examined the association of HGF with stroke in a case/cohort study in the prospective Reasons for Geographical and Racial Differences in Stroke (REGARDS) study, a large longitudinal cohort study of Black and White Americans seeking to elucidate reasons for the racial disparity in stroke mortality. We tested whether racial disparities in stroke could be partially explained by differences in HGF level and if there was effect modification of HGF's association with stroke by race.

2 | METHODS

2.1 | REGARDS study design

The REGARDS study has been described in detail previously [20]. In brief, the study sought to elucidate reasons for regional and racial differences in stroke risk and enrolled 30,239 Black (42%) and White (58%) Americans aged 45 years and older from 2003 to 2007. Participants were 45% male and 55% female, 56% from the 8 states in the Stroke Belt region, and 44% from the other 40 contiguous states. Following verbal consent, medical history, including cardiovascular risk factors, was collected by computer-assisted telephone interview. Thereafter, subjects participated in an in-home examination that included height, weight, and blood pressure (BP) measurements, a resting electrocardiogram (ECG), medication inventory, collection of fasting blood and urine samples, and retrieval of written informed consent. Methods were approved by the institutional review boards of all participating institutions.

2.2 | Covariate definitions

Prevalent stroke was based on a self-report of a physician's diagnosis. BP was the average of 2 seated BP measurements. Hypertension was defined as systolic BP $\geq$ 140 mm Hg, diastolic BP $\geq$ 90 mm Hg, or self-reported physician diagnosis of hypertension with antihypertensive use. Diabetes was defined as fasting glucose $\geq$ 126 mg/dL (or glucose $\geq$ 200 mg/dL among those not fasting or self-reported use of diabetes medications). LVH was defined by ECG criteria [21]. Atrial fibrillation was defined by self-reporting of a physician's diagnosis or presence on ECG. Prebaseline cardiovascular disease was classified as a self-reported physician diagnosis of peripheral arterial disease, myocardial infarction (MI), bypass, angiography, or stenting, or as evidence of prior MI on ECG. Income, education level, warfarin, aspirin, antihypertensive medication, and statin use were based on self-report. Body mass index was calculated from height and weight.

2.3 | Stroke ascertainment

The outcome was the first ischemic stroke through September 1, 2011, with a mean follow-up of 5.2 years (median, 5.5; IQR, 4.0-6.9). Participants were contacted every 6 months via telephone to obtain vital and stroke-free status. Medical records were obtained in the case

of death, reported or suspected stroke, or hospitalization for stroke symptoms identified using the Questionnaire for Verifying Stroke-Free Status [22]. Stroke events were defined as acute, persistent (>24 hours) focal symptoms attributable to obstruction or rupture of the arterial system or stroke evident on brain imaging. Stroke events were adjudicated by blinded independent review using a committee of physicians and based on medical records and imaging review [20]. Strokes were classified as ischemic or hemorrhagic, with further classification of ischemic strokes into subtypes (small vessel, large vessel, cardioembolic, and unclassified)

2.4 | Case-cohort study design

We used a case-cohort study design. Cases were 576 participants stroke-free at baseline with incident ischemic stroke during follow-up. The cohort random sample was selected using stratified random sampling across 20 strata based on age (45-54, 55-64, 65-74, 75-84, and ≥85 years), race, and sex. To ensure sufficient representation of high-risk groups [23], sampling fulfilled the desired distribution: 50% Black, 50% women, and age groups 45 to 54 (20%), 55 to 64 (20%), 65 to 74 (25%), 75 to 84 (25%), and ≥85 (10%) years. Statistical analyses were then weighted to account for stratified sampling. Of 1104 participants in the cohort random sample, we excluded 87 with prebaseline stroke.

2.5 | Laboratory methods

Laboratory methods were described in detail [24]. Briefly, fasting baseline blood samples were drawn in the morning using standardized methods, centrifuged to separate plasma and serum, and shipped overnight on ice to the University of Vermont, where they were processed further and stored at −80 °C. HGF was measured in retrieved plasma of the cases of ischemic stroke and the cohort random sample using a Multiplex Immunoassay (Millipore/Luminex); the interassay coefficient of variation (CV) was 2.98% to 9.54%. N-terminal pro-B-type natriuretic peptide (NT pro-BNP) was measured using an electrochemiluminescence immunoassay (Roche Elecsys 2010; Roche Diagnostics; CV < 5%). D-dimer was measured using an immunoturbidometric assay on the STAR analyzer (Diagnostica Stago); the CV was 5% to 17%. C-reactive protein (CRP) was measured using a high-sensitivity particle-enhanced immunonephelometric assay on the BNII nephelometer (N High Sensitivity CRP, Dade Behring Inc); the CV was 2% to 6%. Total cholesterol, high-density lipoprotein (HDL) cholesterol, and triglycerides were measured by colorimetric reflectance spectrophotometry using the Ortho Vitros Clinical Chemistry System 950IRC instrument (Johnson & Johnson Clinical Diagnostics). Interleukin (IL)-6 was measured by ultrasensitive ELISA (Quantikine HS Human IL-6 Immunoassay) with a CV of 6.3%.

2.6 | Statistical analysis

All analyses were completed using SAS software (SAS Institute). As there is little knowledge on the correlates of HGF, for hypothesis formation, baseline characteristics of participants across HGF quintiles were compared using analysis of variance for continuous variables and chi-squared tests for categorical variables. We then determined independent correlates of HGF in the cohort random sample using a weighted multivariable linear regression model including age, race, sex, and age*race interaction term. Variables from Table 1 were retained in the model if they were significantly associated with HGF. The adjusted mean difference in HGF level by each correlate was calculated.

Hazard ratios (HRs) of stroke by HGF quintile were calculated using Cox proportional hazards models for case-cohort studies with weighting to account for study design using an alpha value of .05. Both linear and quintile analyses were planned *a priori*; HGF quintile was calculated using the cohort random sample distribution. Participants without stroke were censored at death or last follow-up before September 11, 2011. Four successive models were built: (1) model 1 included age, race, sex, and age*race interaction term to account for the larger racial disparity in stroke at a younger age [25]; (2) model 2 added Framingham stroke risk factors (systolic BP, presence or absence of antihypertensive medication use, diabetes mellitus, current smoking, cardiovascular disease, atrial fibrillation, and LVH) [26]; (3) model 3 added other baseline characteristics individually associated with HGF (income, aspirin use, and warfarin use); and (4) model 4 added other biomarkers previously associated with stroke in REGARDS (CRP, D-dimer, NT pro-BNP, and IL-6). Interaction terms using log-transformed HGF for HGF*race and HGF*sex were tested in each successive model. Analysis of HGF in relation to ischemic stroke subtypes was evaluated separately using model 2. Subgroup analyses assessing the association of HGF with stroke incidence in White men, White women, Black men, and Black women were performed. Attenuation analysis of the racial disparity in stroke by HGF was planned but was not performed as HGF did not differ by race.

3 | RESULTS

There were 576 incident ischemic strokes over a mean follow-up time of 5.2 (1.9) years (median, 5.5; IQR, 4.0-6.9), and 557 had HGF measured; 23 of the strokes occurred within the randomly sampled subcohort. In the subcohort (N = 1017), 964 had HGF measured. Baseline characteristics of the cohort random sample by HGF quintile are listed in Table 1. The distribution of HGF was positively skewed in both stroke cases and the cohort random sample. Median HGF was 295 (IQR, 209-402) pg/mL in the cohort random sample. Higher HGF was associated with male sex, older age, lower income, presence of atrial fibrillation, cardiovascular disease, self-reported use of warfarin, aspirin, and antihypertensive medication use. Notably, there was no association of HGF with race. Accounting simultaneously for all observed correlates of HGF, only older age, male sex, current smoking, and warfarin use were independently associated with higher HGF concentration (Table 2). In this model, HGF concentration was 56 pg/mL lower in Black than White participants, but this difference was not statistically significant (95% CI, −156 to 112).

TABLE 1 Baseline characteristics by hepatocyte growth factor level in the cohort random sample.

Characteristic HGF (pg/dL)	Quintile 1, %[a] 13-191	Quintile 2, %[a] 192-257	Quintile 3, %[a] 258-333	Quintile 4, %[a] 334-435	Quintile 5, %[a] 435-10,347
Male gender	41	47	46	57	59
Black race	53	48	50	49	47
Stroke Belt region	36	34	33	38	35
Age (y)	65 (12)	66 (12)	67 (11)	68 (12)	72 (12)
Using antihypertensive medications	55	52	65	60	69
Current smoking	11	10	15	18	18
Diabetes mellitus	14	20	21	22	24
Cardiovascular disease	19	14	23	27	35
Atrial fibrillation	5	7	11	8	13
Left ventricular hypertrophy	13	5	9	9	11
Systolic blood pressure (mm Hg) mean (SD)	127 (17)	126 (16)	128 (16)	130 (18)	130(17)
Income, per y					
0-75,000	71	66	72	69	70
>75,000	21	15	16	14	7
Refused	13	13	10	12	21
Education, % college or above	36	36	38	32	33
Aspirin use	37	39	51	44	41
Warfarin use	<1	2	3	4	6
Statin use	27	30	31	34	33
Body mass index > 30	34	36	36	37	33
Cholesterol (mg/dL), mean (SD)					
Total	192 (34.8)	189 (39.6)	189 (40.8)	188 (42.4)	184 (36.3)
LDL	115 (31.0)	109 (32.1)	112 (36.4)	113 (35.9)	107 (32.4)
HDL	53 (16.6)	53 (18.1)	53 (16.0)	50 (14.9)	51 (17.4)
NT pro-BNP (pg/mL), median (IQR)	73 (34-169)	67(33-135)	75(33-174)	79 (39-224)	129 (52-303)
D-dimer (μg/mL), mean (SD)	3.5 (2.6)	3.4 (2.3)	3.8 (2.8)	4.0 (2.5)	5.1 (3.3)
C-reactive protein (mg/L), mean (IQR)	2.08 (0.92-4.70)	1.66 (0.79-3.83)	1.94 (0.88-4.59)	2.52 (1.20-5.25)	2.60 (1.02-6.48)
IL-6, mean (SD)	3.52 (2.58)	3.36 (2.29)	3.79 (2.84)	3.99 (2.47)	5.09 (3.34)

HDL, high-density lipoprotein; HGF, hepatocyte growth factor; IL, interleukin; LDL, low-density lipoprotein; NT pro-BNP, N-terminal pro-B-type natriuretic peptide.
[a]Unless otherwise specified.

Almost 30% of the strokes occurred among participants in the highest quintile of HGF compared with about 15% in the lowest quintile. In Cox proportional hazard models, each SD increment of log-transformed HGF increased risk of stroke by 30% (HR, 1.30; 95% CI, 1.00-1.70) after adjusting for other stroke risk factors (Table 3). Participants with HGF in the highest quintile had more than a doubling of incident stroke risk (HR, 2.12; OR, 1.31-3.41) compared with those in the lowest quintile. Neither adding other baseline characteristics associated with HGF to the model (model 3) nor adding other stroke biomarkers (model 4: CRP, D-dimer, NT pro-BNP, and IL-6) meaningfully altered the association of HGF with stroke. We assessed for influential outliers by removing participants within the top and bottom 2.5% of HGF, which did not materially affect the HR (data not shown).

HRs of stroke for log-transformed HGF are shown in Figure. The HRs were similar across Black and White men and women.

In the evaluation of ischemic stroke subtypes (Table 4), there were relatively small numbers of participants with each stroke subtype. There was no statistically significant trend across quintiles for any stroke subtype. HGF in the fifth quintile purported the highest risk of stroke across all stroke subtypes, with small vessel and unclassified strokes showing a doubling of stroke risk in the fifth vs first quintile.

TABLE 2 Difference in hepatocyte growth factor by baseline characteristics in a multivariable model.

Baseline characteristic	Difference in HGF (pg/mL)	95% CI
Age, per 10 y	26	9-44
Male sex	60	29-94
Black race	−56	−156 to 112
Cardiovascular disease	27	−7 to 64
Atrial fibrillation	−3	−54 to 51
Systolic blood pressure, per 10 mm Hg	−5	−43 to 38
Antihypertensive medication use	32	0-66
Diabetes	38	5-73
Current smoking	76	29-131
LVH	−4	−45 to 43
Aspirin use	−19	−45 to 8
Warfarin use	109	10-236
Income > $75,000/y	−49	−89 to −3

The multivariable model includes race, Framingham stroke risk factors, and additional Table 1 variables associated with HGF (analysis of variance $P < .05$).

For reference, the median HGF was 294.7 (IQR, 208.7-401.7) pg/mL.

HGF, hepatocyte growth factor; LVH, left ventricular hypertrophy.

4 | DISCUSSION

In this large population study of Black and White people residing in the United States, incident ischemic stroke risk increased with higher baseline HGF, with a doubling of risk in participants with higher HGF that was minimally impacted by adjustment for other stroke risk factors, including novel biomarkers. Results were robust across race and sex subgroups. This considerable effect size is comparable with the effect of smoking or hypertension for incident stroke. It is notable that there was little to no change in the association after adjusting for other stroke risk factors, including traditional and nontraditional risk factors. Though obesity and dyslipidemia are known correlates of HGF [9,15], neither were related to stroke (unpublished data) or HGF level in REGARDS. HGF might represent a unique biology from these risk factors that are relevant to stroke. Despite being a significant risk marker for the development of stroke, HGF concentration did not differ by race and its association with incident stroke did not differ by race, so it does not represent pathways related to race disparities affecting Black individuals.

These findings support prior results of 2 other prospective studies examining HGF and stroke. In the WHI, there was an increased risk of stroke in postmenopausal women with HGF in the highest quartile (HR, 1.39; 95% CI, 1.04-1.85 in a multivariable model) [18] and no difference in HGF by race. In MESA, higher HGF was associated with incident ischemic stroke (HR, 1.14; 95% CI, 1.00-1.31 per each SD increase of HGF in a multivariable model), also showing no difference by race [19] despite identifying some ethnicity-specific gene mutations, which may influence HGF levels [27]. Though these prior studies lacked power to study Black participants separately, our study extends these findings to both Black and White individuals and men and women in a large nationally representative cohort.

There are several potential mechanisms for an association between higher HGF levels and incident stroke. HGF level increases in response to tissue injury and exerts proangiogenic, anti-inflammatory, and antifibrotic effects on endothelial and smooth muscle cells. It may be that preclinical stroke and vascular damage cause upregulation of HGF in response. Other previous work suggested higher levels predict a poor prognosis after ischemic stroke [14], suggesting a correlation between stroke severity/extent of vascular damage and higher HGF. A recent study of vascular biomarkers and magnetic resonance imaging markers of small vessel disease in middle-aged stroke-free individuals supports this hypothesis, showing that HGF was associated with larger cerebrospinal fluid volumes [28], a marker of brain atrophy, even in the absence of overt stroke.

Previous work suggested that HGF may play a more direct role in some subtypes of stroke. HGF increases in response to noncerebral atherosclerosis, and HGF is associated with heart failure severity and mortality [12,13]. Increased circulating levels of HGF and increased HGF expression in atherosclerotic carotid plaques suggest that HGF may also have deleterious effects on the plaque microenvironment,

TABLE 3 Hazard ratio (95% CI) of incident stroke by baseline hepatocyte growth factor quintile.

Model	Quintile 2 vs 1	Quintile 3 vs 1	Quintile 4 vs 1	Quintile 5 vs 1	*P* value for trend
Model 1	1.11 (0.74-1.66)	1.30 (0.88-1.93)	1.19 (0.79-1.78)	1.94 (1.31-2.88)	.008
Model 2	1.47 (0.71-2.38)	1.55 (0.98-2.46)	1.37 (0.85-2.23)	2.12 (1.31-3.41)	.04
Model 3	1.48 (0.91-2.41)	1.49 (0.93-2.38)	1.34 (0.82-2.18)	2.00 (1.22-3.26)	.09
Model 4	1.68 (1.02-2.77)	1.65 (1.02-2.68)	1.23 (0.74-2.05)	2.12 (1.30-3.47)	.02

Model 1: adjusted for age, sex, race, and age*race interaction.

Model 2: model 1 + Framingham stroke risk factors (use of antihypertensive medication, systolic blood pressure, diabetes, left ventricular hypertrophy, cardiovascular disease, atrial fibrillation, and smoking).

Model 3: model 2 + income, aspirin use, and warfarin use.

Model 4: model 3 + D-dimer, N-terminal pro-B-type natriuretic peptide, C-reactive protein, and interleukin-6.

Hepatocyte growth factor quintiles: 13-191, 191-257, 257-333, 333-435, and 435-10,347 pg/mL.

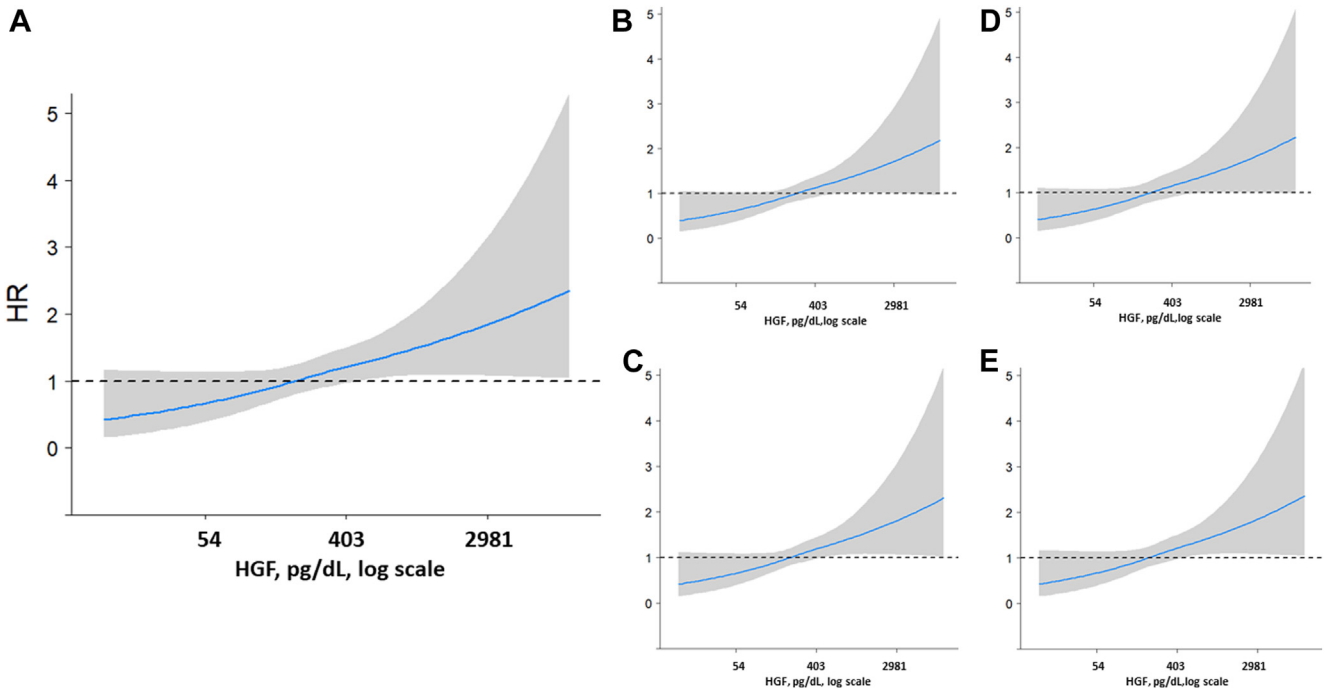

FIGURE Hazard ratio (HR) (blue line) and 95% CI (gray shading) of stroke by baseline hepatocyte growth factor (HGF) level in (A) the entire cohort, (B) Black women, (C) Black men, (D) White women, and (E) White men. HR is anchored around the median value of HGF and log-transformed.

destabilizing and leading to increased symptoms in patients with carotid artery atherosclerosis [29]. Though based on very small numbers, our analyses suggest that higher baseline HGF may be especially associated with small vessel strokes. This preliminary finding supports a hypothesis that HGF is a marker for small vessel disease as a causative pathway rather than cardiac dysfunction. This is also supported by the lack of confounding by NT pro-BNP, a biomarker of heart failure. We did not observe an association with large vessel stroke subtypes in this study; however, the small number of strokes in this subtype limited the power to detect such an association if it exists.

Interestingly, biomarkers previously associated with stroke (CRP [30,31], D-dimer [32], NT pro-BNP [23], and IL-6 [33]) were all positively correlated with HGF, but adjustment for these did not significantly attenuate the association of HGF with stroke. The lack of confounding by these traditional and nontraditional stroke biomarkers suggests these biomarkers represent different biology in the pathogenesis of stroke, and addition of HGF may have utility to better classify stroke risk.

TABLE 4 Hazard ratio (95% CI) of incident stroke subtypes by baseline hepatocyte growth factor quintile.

Stroke subtype	Quintile 1	Quintile 2	Quintile 3	Quintile 4	Quintile 5
All strokes	*N* = 85	*N* = 95	*N* = 114	*N* = 99	*N* = 164
	Reference	1.47 (0.71-2.38)	1.55 (0.98-2.46)	1.37 (0.85-2.23)	2.12 (1.31-3.41)
Cardioembolic	*N* = 18	*N* = 20	*N* = 14	*N* = 20	*N* = 37
	Reference	1.29 (0.51-3.24)	0.79 (0.31-2.03)	1.18 (0.47-2.97)	1.50 (0.63-3.59)
Large vessel	*N* = 14	*N* = 14	*N* = 11	*N* = 15	*N* = 16
	Reference	1.26 (0.62-0.57)	0.88 (0.42-1.83)	1.19 (0.57-2.46)	1.45 (0.72-2.92)
Small vessel	*N* = 8	*N* = 13	*N* = 19	*N* = 18	*N* = 29
	Reference	1.53 (0.77-3.07)	1.27 (0.64-2.52)	1.53 (0.77-3.05)	2.17 (1.12-4.19)
Unclassified/	*N* = 36	*N* = 40	*N* = 58	*N* = 42	*N* = 71
other	Reference	1.46 (0.85-2.52)	1.53 (0.91-2.57)	1.26 (0.73-2.17)	2.08 (1.23-3.53)

Adjusted for age, sex, race, race*age interaction term, and Framingham stroke risk factors (use of antihypertensive medication, systolic blood pressure, diabetes, left ventricular hypertrophy, cardiovascular disease, atrial fibrillation, and smoking).

N indicates the number of strokes in each group.

P value for trend > .10 for all stroke subtypes.

Prior animal model research showing improved cardiac outcomes with higher HGF levels after MI [6] and treatment of animals with HGF [34] following induced MI suggested HGF had potential therapeutic effect immediately following MI. HGF level at the time of stroke is associated with worse stroke severity and outcome after stroke [14,15], which may be due to a greater degree of endothelial damage in these patients, resulting in a surge of HGF in response. HGF could have therapeutic value in stroke as well. However, given possible implications in pathogenesis in carotid plaque destabilization and HGF's pleiotropic nature with many other target tissues (antagonism of HGF has shown some promise in cancer therapeutics [7]), more basic research into the mechanism of the association of HGF with stroke is needed. Though our results are compelling for a strong association of HGF with incident stroke, the clinical potential of measuring HGF levels remains unknown.

Strengths of this study include its prospective design, well-defined baseline variables, outcome ascertainment, large numbers of ischemic stroke events, and power to detect differences in associations between Black and White participants. Limitations include relatively few strokes in specific subtypes, limiting power to detect differences. Stroke subtyping was also not always possible due to variability in clinical evaluation at the large number of hospitals where participants were seen. However, even with this limitation, we are confident in the classifications made and have identified suggestive differences in the association among subtypes, helping to suggest the driving mechanism for the overall association of HGF with stroke. Additionally, HGF was measured only once at baseline, in some cases years before development of incident stroke. This indicates that the observed association might underestimate the strength of the true association and that serial measurement might reveal this. Other limitations are the reliance on patient self-reporting baseline conditions and lack of time-varying medication use, including other antiplatelet medication use. It is also possible that secular trends might impact findings if enrollment was more recent.

5 | CONCLUSION

Higher HGF was associated with incident stroke in REGARDS, a nationally representative cohort of Black and White Americans. Baseline HGF did not differ by race, and there was no interaction between race and HGF on incident stroke. Therefore, HGF did not mediate the excess stroke risk in Black Americans. The association of HGF with stroke may be strongest in small vessel strokes, but we lacked power in stroke subgroups to explore this fully. Further study of the mechanisms of this association is of high priority.

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AUTHOR CONTRIBUTIONS

S.G., N.Z., and M.C. contributed to the concept and design of the study, analysis and/or interpretation of data, and critical writing or revising the intellectual content. I.K. contributed to the analysis and/or interpretation of data. S.J. contributed to the concept and design of the study and analysis and/or interpretation of data. T.P. contributed to the analysis and/or interpretation of data and critical writing or revising the intellectual content. G.H. contributed to the concept and design of the study, interpretation of data, and critical writing or revising the intellectual content. All authors read and approved the final version of the manuscript.

RELATIONSHIP DISCLOSURE

The authors have no conflicts of interest to disclose.

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