Hepatocyte Growth Factor and Left Ventricular Geometry in End-Stage Renal Disease

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Abstract—Hepatocyte growth factor is a pleiotropic cytokine with cardioprotective properties. Its serum concentration is markedly raised in end-stage renal disease. This study assessed the relation of hepatocyte growth factor (HGF) with left ventricular mass and geometry in end-stage renal disease. Serum HGF measurements and echocardiographic studies were performed in 185 patients receiving hemodialysis. Patients with serum HGF above the median (1.85 ng/mL) had more frequent cardiovascular complications. This cytokine was directly related to mean left ventricular wall thickness ($r=0.23$, $P=0.002$) and relative wall thickness ($r=0.25$, $P=0.0001$); a multivariate analysis showed that this relation was independent of other risk factors. Accordingly, the prevalence of left ventricular concentric geometry (either concentric left ventricular hypertrophy or remodeling) was much higher ($n=49$, 53%) among patients with HGF values above the median that in those with values $\leq$1.85 ng/mL ($n=31$, 34%). Furthermore, the risk for left ventricular concentric geometry was higher in patients with HGF values above the median (odds ratio, 2.57; 95% CI, 1.33 to 4.98; $P=0.005$), and multiple logistic regression analysis confirmed that this association was independent of other risk factors. In patients receiving hemodialysis, elevated serum HGF is associated with concentric left ventricular geometry. This is consistent with reports that link this cytokine to arterial remodeling and survival in patients with end-stage renal disease and suggests that it is part of a counterregulatory response aimed at attenuating cardiovascular damage in this high-risk population. (*Hypertension*. 2003;41:88-92.)

Key Words: growth substances ■ renal disease ■ remodeling ■ hypertrophy

The mesenchyme-derived cytokine hepatocyte growth factor (HGF) has a wide range of biological effects spanning from liver regeneration to protection and/or repair of various organs including the kidney and the cardiovascular system.1–4 The role of HGF in the cardiovascular system has been recently investigated. Specifically, HGF appears to be particularly important in improving endothelial function and attenuating tissue fibrosis.5 HGF and its transmembrane tyrosine kinase receptor, c-met, are present in the human heart, and their production is increased after myocardial infarction.6 Furthermore, enhanced secretion of cardiac HGF from the infarcted areas is associated with reduced ventricular enlargement and improved cardiac function in humans.7 Therefore this cytokine, which is a growth factor that modulates endothelial and myocardial repair,8–10 may contribute to myocardial remodeling in situations of cardiovascular stress.

Renal failure is associated with a high prevalence of left ventricular hypertrophy (LVH), which is the strongest predictor of death in end-stage renal disease (ESRD) patients.11,12 Therefore, ESRD is an interesting natural model to explore the relation between serum HGF and the cardiovascular system. Studies have reported that this cytokine is markedly raised in patients with ESRD13–16 and may be considered a novel vascular modulator in humans.17,18 Of note, we previously reported that serum HGF in ESRD correlated with carotid intima-media thickness, thus behaving as a marker of atherosclerosis.19 This link is of particular interest because we have also shown that serum HGF is an independent predictor of death in uremic patients.19 Because serum HGF interferes with mechanisms regulating myocardial trophism and plasticity,7 it may be also involved in ventricular remodeling in patients with ESRD. We therefore thought to investigate the relation between serum HGF and echocardiographic parameters of the left ventricle in patients receiving long-term hemodialysis.

Methods

The protocol was approved by our institutional committee and was in accordance with institutional guidelines. Each participant gave informed consent.
Study Population

Patients receiving hemodialysis patients (n=185; 81 women) with ESRD on regular dialysis treatment (RDT) for at least 6 months (median duration, RDT 43 months; interquartile range, 19 to 110 months) were eligible.

Patients were anuric (24-hour urine volume <200 mL/d) and were treated 3 times weekly with standard bicarbonate dialysis. Dry weight was targeted to achieve a normotensive edema-free state. Average fractional urea clearance (Kt/V) was 1.21 ± 0.27. Twenty-three patients were diabetic, 74 were smokers, and 88 had had cardiovascular events (ECG-documented angina, myocardial infarction, transient ischemic attack, stroke, or peripheral vascular disease). Ninety-nine patients were treated with erythropoietin and 67 received antihypertensive medication. Patient characteristics are shown in Table 1.

Blood Pressure

Predialysis blood pressure was measured 3 times per week and averaged in the 4 weeks preceding the study. 20

Laboratory

Blood was sampled midweek between two dialyses after 20 to 30 minutes of resting in a semirecumbent position. Samples were collected in prechilled EDTA tubes and centrifuged within 30 minutes at 4°C. Plasma was stored at −80°C until analysis. Fasting blood was drawn for serum HGF, calcium, phosphate, total cholesterol, triglycerides, albumin, hemoglobin, homocysteine, 21 and C-reactive protein (CRP).

HGF was determined by solid-phase ELISA (R&D Systems Europe). Intra-assay and interassay precision of this method are 5.6% and 7.0%, respectively. The normal range for serum HGF in healthy individuals (n=82) is 0.319 to 1.475 ng/mL.

Echocardiography

Echocardiographic studies were performed according to American Society of Echocardiography recommendations on a nondialysis day, within 2 hours after blood sampling, by an observer unaware of biochemical results. Left ventricular mass (LVM) was calculated according to the Devereux formula and specifically indexed to body surface area (BSA) according to the Devereux formula. Concentric and eccentric left ventricular geometry was established according to Ganau et al. 24 Mean wall thickness (MWT) was calculated as (interventricular septum + posterior wall thickness)/2.

Statistical Analysis

Data are reported as mean±SD and median and interquartile range or percent frequency. Comparisons between groups were made by t test, Mann-Whitney test, or χ² test. Relations between paired parameters were analyzed by the least-squares method, with and without outliers. Significant outliers were identified by Grubbs test. 25 Independent relations between HGF and LVM and geometry were assessed by multivariate models based on serum HGF and on
Similarly, time on RDT and hemoglobin were unrelated to serum HGF did not correlate with LVMI ($r=0.08$, $P=0.29$).

Because of the opposite correlations of serum HGF with the cytokine were inversely related to LVEDD ($r=-0.15$, $P=0.04$). Because of the opposite correlations of serum HGF with the muscular and cavitary components of the left ventricle (direct correlation with MWT and inverse correlation with LVEDD), serum HGF did not correlate with LVMI ($r=0.08$, $P=0.29$).

Similarly, time on RDT and hemoglobin were unrelated to traditional and emerging cardiovascular risk factors in dialysis patients (age, male sex, previous cardiovascular events, smoking, diabetes, systolic pressure, antihypertensive therapy, albumin, hemoglobin, cholesterol, calcium, phosphate, Kt/V, homocysteine, and CRP). Independent variables were identified by stepwise approach. Potential confounding effect of covariates ($P<0.10$) in the HGF groups (Table 1) was evaluated by introducing such covariates into the final models.

Data are expressed as regression coefficient ($\beta$) or as odds ratio and 95% CI. Analysis was done with SPSS for Windows 9.0.1.

**Results**

In the majority of patients receiving dialysis (135 of 185 patients, that is, 73%), serum HGF (median, 1.85 ng/mL; interquartile range, 1.42 to 2.53 ng/mL; mean±SD, 2.68±3.60 ng/mL) was above the upper limit of the normal range (cutoff, 1.47 ng/mL).

On echocardiography, concentric geometry was the most frequent pattern [n=80, 43.2% (concentric LVH: n=68; concentric remodeling: n=12)], followed by eccentric LVH (n=69, 37.3%). Only a minority of patients showed a normal LVM and LV geometry (n=36, 19.5%). As shown in Table 1, patients with serum HGF above the median value (cutoff $>1.85$ ng/mL) more frequently had had cardiovascular complications than those with serum HGF below or equal to the median. Serum CRP was higher in the patients with HGF $>1.85$ ng/mL. Notably, patients with serum HGF above the median value had higher MWT and RWT ($P=0.005$, for both) and tended to have lower LVEDD ($P=0.09$) in comparison to those with serum HGF below this threshold. Accordingly, on univariate analysis, serum HGF was directly related to MWT and RWT (Figure, top) and showed a progressive rise from the first to the fourth HGF quartile (Figure, bottom). Furthermore, the plasma levels of this cytokine were inversely related to LVEDD ($r=-0.15$, $P=0.04$).

Because of the opposite correlations of serum HGF with the muscular and cavitary components of the left ventricle (direct correlation with MWT and inverse correlation with LVEDD), serum HGF did not correlate with LVMI ($r=0.08$, $P=0.29$). Similarly, time on RDT and hemoglobin were unrelated to echocardiographic data. Data analysis of the LV geometric patterns showed a substantially higher prevalence of LV concentric geometry in patients with serum HGF above the median value (n=49, 53%) in comparison to those below the median value (n=31, 34%) ($P=0.009$). Average predialysis blood pressure values of 2 to 3 months did not improve the relation between blood pressure and echocardiographic parameters.

The associations of serum HGF with the muscular component of left ventricle (MWT) as well as with RWT and LVEDD were confirmed in multiple regression models. In these models, serum HGF was an independent correlate of these echocardiographic parameters (Table 2). Similarly, multiple logistic regression analysis confirmed that the association of serum HGF with LV concentric geometry was independent of other risk factors (Table 3). In this model, patients with serum HGF above the median value had a risk for LV concentric geometry that was 2.57 times (95% CI, 1.33 to 4.98) higher than those with serum HGF below this threshold ($P=0.005$). The administered dialysis dose, as by measured fractional urea clearance (Kt/V), did not influence these links either on multiple linear or logistic regression analysis ($P>0.80$).

**Discussion**

In this study, serum HGF was linked to LV wall thickness and to concentric remodeling and hypertrophy. Importantly, these links were independent of traditional and emerging cardiovascular risk factors as well as of administered dialysis dose.

Consistent with other studies that have reported elevated serum HGF level in patients with chronic renal failure and particularly in those in the end-stage phase, we found that the serum concentration of HGF is considerably increased in patients receiving hemodialysis. HGF has a short half-life and is cleared mainly by the liver. Although the precise mechanism(s) responsible for the high levels of serum HGF in ESRD are still unclear, it appears that this phenomenon does not represent the mere effect of reduced removal of this substance by the diseased kidneys. It has been shown that the synthesis of

Semilogarithmic relations among serum HGF and MWT and RWT (top). Bottom, Average MWT and RWT in patients grouped into HGF quartiles. Symbol “O” in figures (top) identifies the outlier (lg10 HGF=1.56; MWT=1.6 cm and lg10 HGF=1.56; RWT=0.78 cm). Strength of relations among serum HGF and MWT ($r=0.18$, $P=0.02$) and RWT ($r=0.19$, $P=0.01$) was reduced slightly after exclusion of the outlier.
HGF is markedly upregulated in different organ systems in response to various illnesses such as acute renal failure and myocardial infarction. The relation between circulating HGF and atherosclerosis has recently received increased attention. In a population of healthy individuals, increased HGF has been shown to be associated with carotid artery remodeling. These findings are in line with previous observations by our group, which show HGF to correlate with intima-media thickness independently of other risk factors for atherosclerosis. Hence, the association between higher HGF and higher CRP levels is not surprising, the latter being a known marker of atherosclerosis. Arterial and cardiac remodeling proceed in parallel in patients with renal failure. In the present study, we have therefore tested the hypothesis that the association between intimal lesions in the carotid arteries and HGF is also extended to the heart. This question is important because vascular-ventricular coupling may be modulated by various endogenous factors in uremic subjects. In this regard, it is worth noting that whereas endogenous inhibitors of NO synthase, for example, asymmetric dimethylarginine, are associated with arterial and myocardial remodeling, such parallelism was not found in patients with ESRD for other emerging risk factors such as advanced glycation end-products. LVH is indeed a multifactorial phenomenon, and in this study multiple regression analysis showed that HGF explained only in small part the variability in the muscular component of the left ventricle.

However, because of the opposite correlations of serum HGF with the muscular and cavitary components of the left ventricle (direct correlation with MWT and inverse correlation with LVEDD), HGF did not correlate with LVMI. Patients with ESRD stratified according to serum HGF revealed a higher prevalence of previous cardiovascular events in the group with serum HGF above the median value. Individuals who had had cardiovascular events are notoriously at higher risk for further cardiovascular complications. This is consistent with observations made in a prospective cohort study showing that high HGF is independently associated with death in these patients. Of note, however, the association between HGF and echocardiographic parameters of the LV found in the present study was independent of previous cardiovascular events. This indicates that the serum concentration of HGF reflects a continuous, graded process related to the muscular component of the heart and that this process is not confined to high-risk individuals. This direct link between HGF and LV wall thickening may underscore a compensatory mechanism aimed at reducing cardiac fibrosis.

Although the cross-sectional design of our study precludes any conclusion on the nature of this link (causal or noncausal), the biological effects of HGF would therefore suggest that the association between HGF and myocardial hypertrophy reflects a mechanism aimed at attenuating the adverse effects of other risk factors. Whatever the explanation for this intriguing association, our data indicate that HGF may be a marker of concentric geometry, that is, a geometric alteration of the LV, which is strongly associated with all-cause and cardiovascular death in the dialysis population. Moreover, the logistic regression further underscores that serum HGF together with serum cholesterol and albumin are associated with LV concentric hypertrophy. In this respect, there is solid evidence both in animal models of chronic renal failure and in uremic patients that LVH is characterized by a high fibrotic component and that such process is probably a major cause of ventricular dysfunction in ESRD. Hence, establishing whether elevated HGF contributes to attenuate fibrosis is an important question with potential therapeutic implications also in light of recent findings showing therapeutic potential for HGF in myocardial regeneration therapy for heart failure.

### TABLE 2. Multivariate Analysis of Mean Wall Thickness, Relative Wall Thickness, and Left Ventricular End Diastolic Diameter

<table>
<thead>
<tr>
<th>Variable</th>
<th>β</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mean wall thickness; multiple R=0.58, P&lt;0.001</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Albumin</td>
<td>−0.38</td>
<td>0.0001</td>
</tr>
<tr>
<td>Systolic pressure</td>
<td>0.24</td>
<td>0.001</td>
</tr>
<tr>
<td>HGF</td>
<td>0.17</td>
<td>0.008</td>
</tr>
<tr>
<td>Smoking</td>
<td>0.15</td>
<td>0.02</td>
</tr>
<tr>
<td>Antihypertensive therapy</td>
<td>0.14</td>
<td>0.049</td>
</tr>
<tr>
<td>Cholesterol</td>
<td>0.13</td>
<td>0.06</td>
</tr>
<tr>
<td>Previous CV events</td>
<td>0.10</td>
<td>0.16</td>
</tr>
<tr>
<td>CRP</td>
<td>0.05</td>
<td>0.47</td>
</tr>
<tr>
<td>Age</td>
<td>0.02</td>
<td>0.83</td>
</tr>
<tr>
<td>Relative wall thickness; multiple R=0.47, P&lt;0.001</td>
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<td></td>
</tr>
<tr>
<td>Albumin</td>
<td>−0.38</td>
<td>0.0001</td>
</tr>
<tr>
<td>Cholesterol</td>
<td>0.23</td>
<td>0.003</td>
</tr>
<tr>
<td>HGF</td>
<td>0.19</td>
<td>0.007</td>
</tr>
<tr>
<td>Systolic pressure</td>
<td>0.13</td>
<td>0.08</td>
</tr>
<tr>
<td>Antihypertensive therapy</td>
<td>0.08</td>
<td>0.26</td>
</tr>
<tr>
<td>Age</td>
<td>−0.08</td>
<td>0.32</td>
</tr>
<tr>
<td>CRP</td>
<td>0.06</td>
<td>0.38</td>
</tr>
<tr>
<td>Previous CV events</td>
<td>0.06</td>
<td>0.42</td>
</tr>
<tr>
<td>Left ventricular end diastolic diameter; multiple R=0.44, P&lt;0.001</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Male gender</td>
<td>0.40</td>
<td>0.0001</td>
</tr>
<tr>
<td>HGF</td>
<td>−0.19</td>
<td>0.008</td>
</tr>
<tr>
<td>Previous CV events</td>
<td>0.14</td>
<td>0.08</td>
</tr>
<tr>
<td>Age</td>
<td>0.12</td>
<td>0.10</td>
</tr>
<tr>
<td>CRP</td>
<td>−0.08</td>
<td>0.25</td>
</tr>
<tr>
<td>Antihypertensive therapy</td>
<td>0.05</td>
<td>0.45</td>
</tr>
</tbody>
</table>

Data are expressed as standardized regression coefficients (β) and P values.

### TABLE 3. Multiple Logistic Regression Analysis of LV Concentric Geometry

<table>
<thead>
<tr>
<th>Variable</th>
<th>Unit of Increase</th>
<th>Odds Ratio (95% CI)</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>Albumin</td>
<td>1 g/L</td>
<td>0.90 (0.83–0.97)</td>
<td>0.005</td>
</tr>
<tr>
<td>HGF</td>
<td>≤1.85 ng/mL</td>
<td>1*</td>
<td>0.005</td>
</tr>
<tr>
<td></td>
<td>&gt;1.85 ng/mL</td>
<td>2.57 (1.33–4.98)</td>
<td>0.02</td>
</tr>
<tr>
<td>Antihypertensive therapy</td>
<td>2.23 (1.14–4.37)</td>
<td>0.02</td>
<td></td>
</tr>
<tr>
<td>Cholesterol</td>
<td>1 mmol/L</td>
<td>1.32 (1.02–1.69)</td>
<td>0.03</td>
</tr>
<tr>
<td>Age</td>
<td>1 year</td>
<td>1.98 (0.96–1.01)</td>
<td>0.13</td>
</tr>
<tr>
<td>Previous CV events</td>
<td>1.11 (0.55–2.26)</td>
<td>0.77</td>
<td></td>
</tr>
<tr>
<td>CRP</td>
<td>1 mg/L</td>
<td>1.00 (0.99–1.01)</td>
<td>0.83</td>
</tr>
</tbody>
</table>

Data are expressed as odds ratio and 95% confidence interval.

*Reference group.
The results of the present study demonstrate an association between serum HGF and concentric LVH and remodeling in uremic patients.

Perspectives

These results are consistent with previous data showing that HGF is linked to arterial remodeling and survival in patients receiving hemodialysis and further suggest that this cytokine may be part of a counterregulatory response that attenuates cardiovascular damage in the high-risk population of patients with ESRD. The cardiovascular mortality rate in uremic patients is high, and LVH control, particularly in patients with concentric LVH, is considered a priority. Therefore, HGF and its c-met receptor may become a target for therapeutic intervention.

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