

Circulation

JOURNAL OF THE AMERICAN HEART ASSOCIATION



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Circulation 2003;107;524-530; originally published online Jan 20, 2003;
DOI: 10.1161/01.CIR.0000048183.37648.1A

Circulation is published by the American Heart Association, 7272 Greenville Avenue, Dallas, TX 75214

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Prognostic Significance of Angiogenic Growth Factor Serum Levels in Patients With Acute Coronary Syndromes

Christopher Heeschen, MD; Stefanie Dimmeler, PhD; Christian W. Hamm, MD; Eric Boersma, MD; Andreas M. Zeiher, MD; Maarten L. Simoons, MD; on Behalf of the CAPTURE (c7E3 Anti-Platelet Therapy in Unstable REfractory angina) Investigators

Background—In patients with acute coronary syndromes, compensatory processes are initiated, including angiogenesis and endothelial regeneration of ruptured or eroded plaques. Angiogenic growth factors like vascular endothelial growth factor (VEGF), hepatocyte growth factor (HGF), and basic fibroblast growth factor (bFGF) are upregulated during ischemia. However, it is unknown whether their serum levels are related to clinical outcome.

Methods and Results—We measured VEGF, HGF, and bFGF levels in 1090 patients with acute coronary syndromes. Angiographic evaluation was performed at baseline as well as death, and nonfatal myocardial infarctions were recorded during 6-month follow-up. HGF and VEGF, but not bFGF, were significantly and independently associated with the patients' outcome. Patients with elevated VEGF serum levels suffered from adverse outcome (adjusted hazard ratio, 2.50 [1.52 to 4.82]; $P=0.002$). VEGF elevation was associated with evidence of ischemia and was a significant predictor of the effect of glycoprotein IIb/IIIa inhibition. In contrast, patients with high HGF levels had a significantly lower event rate compared with patients with low HGF levels (adjusted hazard ratio, 0.33 [0.21 to 0.51]; $P<0.001$). HGF levels did not correlate with evidence of ischemia and did not predict the effect of abciximab. Intriguingly, however, HGF levels significantly correlated with angiographically visible collateralization of the target vessel (22.4% versus 10.5%; $P<0.001$).

Conclusions—The angiogenic growth factors VEGF and HGF are independent predictors of the patients' prognosis in acute coronary syndromes. Whereas VEGF elevation correlated with the evidence of myocardial ischemia and indicated an adverse outcome, HGF elevation was independent of ischemia and associated with improved collateralization as well as a favorable prognosis. (*Circulation*. 2003;107:524-530.)

Key Words: angina ■ angiogenesis ■ prognosis

In recent years, biochemical markers have contributed to a better understanding of the underlying pathophysiology in patients with acute coronary syndromes. Markers of myocardial necrosis, most notably cardiac troponins, have become valuable tools for risk stratification and optimizing treatment strategies.¹⁻⁵ During the consecutive repair phase after onset of an acute coronary syndrome, compensatory processes are initiated, including the formation of collateral circulation as well as endothelial regeneration of ruptured or eroded plaques.⁶ A variety of potent angiogenic growth factors have been identified to stimulate neovascularization of ischemic tissue.^{7,8} Among these, vascular endothelial growth factor (VEGF), hepatocyte growth factor (HGF), and basic fibroblast growth factor (bFGF) have been experimentally shown to improve tissue perfusion when administered exogenously into ischemic tissue.⁹⁻¹¹ In addition, both VEGF and HGF have been shown to be upregulated endogenously after myocardial infarction.^{12,13}

However, it is unknown how serum levels of these angiogenic growth factors relate to clinical outcome in patients with acute coronary syndromes. In a previous study including patients with myocardial infarction, baseline VEGF serum levels were found to be elevated in patients with preinfarction angina, a clinical feature associated with increased cardiac risk, whereas HGF serum levels did not differ between patients with and without preinfarction angina.¹⁴ Other studies for HGF revealed findings that are consistent with a protective effect of HGF in patients with unstable coronary heart disease.¹⁵⁻¹⁷ To investigate the prognostic significance of angiogenic growth factors in patients with acute coronary syndromes, we measured VEGF, HGF, and bFGF serum levels in patients with acute coronary syndromes enrolled in the CAPTURE trial (c7E3 Anti Platelet Therapy in Unstable Refractory angina). The CAPTURE study was designed to assess outcome in patients with refractory angina receiving

Received September 30, 2002; revision received October 17, 2002; accepted October 20, 2002.

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DOI: 10.1161/01.CIR.0000048183.37648.1A

either abciximab or placebo up to 24 hours before scheduled percutaneous transluminal coronary angioplasty.¹⁸

Methods

Patients

The CAPTURE trial enrolled 1265 patients with refractory unstable angina (61% males, aged 61 ± 10 years) between May 1993 and December 1995. All CAPTURE patients had recurrent chest pain at rest associated with electrocardiographic changes during treatment with intravenous heparin and glyceryl trinitrate. All patients underwent coronary angiography before randomization, indicating significant coronary artery disease with a culprit lesion ≥70% suitable for angioplasty. The patients were randomly assigned to abciximab or placebo; treatment was initiated within 2 hours of randomization. Heparin was administered from before randomization until at least 1 hour after the percutaneous transluminal coronary angioplasty procedure and adjusted to achieve an activated partial thromboplastin time between 2.0 and 2.5 times normal. For all patients, coronary interventions were scheduled between 18 and 24 hours after beginning study treatment. Primary end points of our study were mortality and nonfatal myocardial infarction during the 6-month follow-up period.¹⁸

Coronary Angiogram

All angiograms were assessed centrally by an Angiographic Committee at Cardialysis, Rotterdam, the Netherlands, with respect to TIMI flow,¹⁹ characteristics of the culprit lesion according to the AHA/ACC grading system, thrombus formation, and collateralization. Presence of thrombus was strictly defined as filling defect or haziness without calcification near the lesion visible on at least 2 orthogonal views or an embolus in the distal territory of the related artery. Collateralization was categorized as undetectable (0), minimal (1), partial (2), and complete retrograde filling (3).

Analytical Techniques

Blood samples were collected 8.7 hours (75% CI, 3.6 to 11.3) after onset of symptoms. HGF,¹⁷ bFGF,²⁰ and VEGF²¹ were measured by ELISA (all from R&D systems). Detection limit for the respective assays was 0.04 µg/L, 0.05 ng/L, and 0.005 pg/L. Intra-assay variation among triplicates was 7.5% for HGF, 8.2% for bFGF, and 6.6% for VEGF. It should be noted that HGF levels have been shown to increase remarkably within 10 minutes after intravenous administration of heparin.¹⁷ For quantification of troponin T (TnT), a one-step enzyme immunoassay based on electrochemiluminescence technology (Elecsys 2010; Roche Diagnostics) was used.²² The detection limit of this assay is 0.01 µg/L, and the diagnostic threshold level is 0.10 µg/L. C-reactive protein (CRP) was measured by N latex CRP mono tests, performed on a Behring BN II Nephelometer (Behring Diagnostics).²³ The detection limit of the assay is 0.2 mg/L, and a calculated diagnostic threshold level of 10.0 mg/L was used.²

Statistical Methods

To distinguish between patients with different degrees of cardiac risk, an exploratory data analysis was chosen. Patients were categorized in quartiles according to the marker concentration. For each of the 4 time points (24 hours, 72 hours, 30 days, and 6 months), a Cox proportional-hazards regression analysis for the combined end points death and nonfatal myocardial infarction was performed and patients in the first quartile served as reference. Receiver operating characteristics curve analyses over the dynamic range of the assays was used to identify the threshold level providing highest predictive value for risk-stratifying patients with acute coronary syndromes. The impact of elevation of biochemical markers on patients' outcome was evaluated using a Cox proportional-hazards regression model, adjusting for baseline prognostic factors (eg, electrocardiographic findings, cardiac risk factors, age, and sex), use of heparin, and randomized treatment.²⁴

All results for continuous variables are expressed as medians with 95% confidence intervals. Comparisons between groups were analyzed by *t* test (two-sided) or ANOVA for experiments with more than 2 subgroups. Post-hoc range tests and pairwise multiple comparisons were performed with the *t* test (two-sided) with Bonferroni adjustment. Comparison of categorical variables was generated by the Pearson χ^2 test. *P* < 0.05 was considered statistically significant.

Results

Serum samples were available in 1090 of 1265 patients (86.2%). The baseline characteristics for the substudy population were not different from the total study population. The reduction of cardiac events in the abciximab group of the substudy population was comparable to the entire CAPTURE study population.

Elevated HGF Levels Predict Favorable Outcome

Overall HGF concentration did not correlate with TnT levels (*R* = 0.002; *P* = 0.95) or CRP levels (*R* = 0.032; *P* = 0.30). Patients were stratified into quartiles according to their measured HGF levels, as follows: HGF 1, <2.50 µg/L (n = 260); HGF 2, 2.50 to 4.70 µg/L (n = 278); HGF 3, 4.71 to

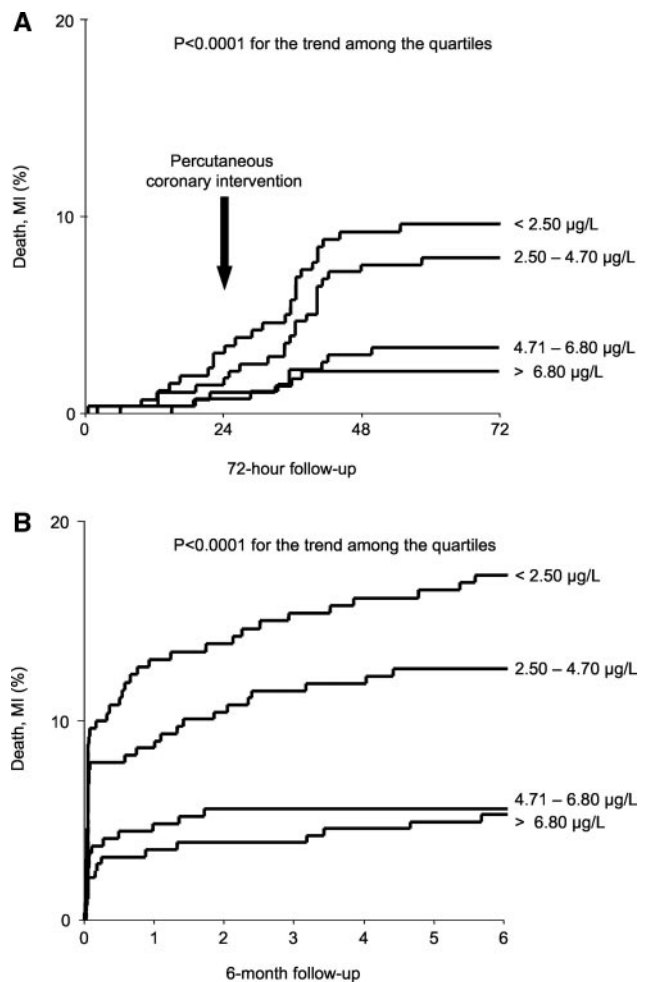


Figure 1. Association between HGF serum levels and cardiac event rates. Patients in the upper 2 quartiles had a significantly better outcome at 72-hour follow-up compared with patients in the first quartile (A). Event rate curves continued to diverge during the subsequent 6-month follow-up (B).

TABLE 1. Baseline Characteristics According to HGF, VEGF, and bFGF Status

	HGF Low	HGF High	VEGF Low	VEGF High	bFGF Low	bFGF High
No.	538	552	555	535	813	277
Male, %	72.3	70.1	69.4	72.0	70.6	74.1
Age, y	62.1±10.1	60.4±10.6	59.7±10.3	62.6±10.5	60.1±10.2	64.7±10.4
History, %						
Angina >4 weeks	57.3	55.6	54.5	58.6	55.4	60.3
Infarction <30 days	21.0	18.0	18.6	20.5	19.6	19.5
Infarction >30 days	19.0	16.3	15.9	19.4	17.7	17.3
PTCA	18.5	16.6	16.1	19.0	17.3	18.4
CABG	3.4	3.5	3.5	3.4	3.7	2.5
Baseline ECG, %						
Transient ST elevation	34.2	33.7	29.1	38.8*	33.6	35.0
ST depression	45.2	44.2	30.9	58.5*	43.2	49.5
T-wave inversion	47.3	54.6	52.5	49.5	50.6	52.3
Risk factors, %						
Diabetes	10.4	9.9	8.2	12.1	10.0	10.5
Hypertension	35.4	38.5	36.2	37.7	36.9	37.2
Current smokers	38.3	42.5	41.2	39.6	40.3	40.8
Medication at baseline, %						
Aspirin	98.1	98.0	98.1	98.1	98.1	98.2
Nitrates IV	98.9	99.6	99.2	99.3	99.6	97.8
β-Blockers	64.0	62.7	63.5	63.2	63.0	64.3
ACE inhibitors	19.3	22.5	20.9	20.8	21.0	20.5
Ca antagonists	52.3	55.4	56.1	51.6	53.9	53.8
Heparin IV, %						
Dose, IU per hour	148.8±35.9	143.6±41.3	146.5±38.7	144.3±38.5	144.9±35.8	150.2±50.4
aPTT, x-fold normal	2.31±0.41	2.25±0.39	2.29±0.42	2.27±0.38	2.25±0.39	2.36±0.48
Troponin T ≥0.1 μg/L, %	35.5	40.6	18.9	57.9*	37.5	41.2
CK-MB mass ≥5.0 μg/L, %	14.7	14.5	8.3	21.3*	13.7	17.3

* $P<0.01$.

PTCA indicates percutaneous transluminal coronary angioplasty; CABG, coronary artery bypass graft; and ACE, angiotensin-converting enzyme.

6.80 μg/L (n=283); and HGF 4, >6.80 μg/L (n=269), respectively. For the initial 24-hour period, the combined end points mortality and nonfatal myocardial infarction tended to be lower in the fourth quartile compared with the first quartile ($P=0.15$) (Figure 1A). For the 72-hour follow-up, including peri-interventional events, the event rates in the third and fourth quartiles were significantly lower compared with the first quartile ($P<0.001$ for both). During 30-day and 6-month follow-up, the event rate curves additionally diverged, and event rates remained significantly lower in the third and fourth quartile ($P<0.001$ for all time points) (Figure 1B). Consistent with these results, receiver operating characteristics curve analysis indicated a threshold level of 4.7 μg/L HGF for maximized predictive value.

Therefore, the study population was dichotomized according to the calculated threshold level of 4.7 μg/L, resulting in 552 patients with elevated HGF levels (50.6%). As depicted in Table 1, there were no significant differences in the baseline characteristics of the 2 groups. Importantly, heparin dose and activity did not differ between patients with high and low HGF serum levels. The adjusted hazard ratios were 0.29 (95% CI, 0.16 to 0.53; $P<0.001$)

for the 72-hour follow-up, 0.30 (0.18 to 0.52; $P<0.001$) for the 30-day follow-up, and 0.33 (0.21 to 0.51; $P<0.001$) for the 6-month follow-up. Six-month cumulative event rates in patients with low HGF levels were 14.9% versus 5.4% for patients with high HGF levels ($P<0.001$). This difference in event rates was not only driven by a higher rate of nonfatal myocardial infarction but also by a significantly higher mortality in patients with reduced HGF serum levels (3.3% versus 0.7%; $P=0.002$). Most notably, if HGF serum levels were below the calculated threshold level, even in the absence of myocardial necrosis as evidenced by normal TnT serum levels, patients suffered from increased cardiac risk (adjusted hazard ratio, 8.20 [2.43 to 27.60]; $P<0.001$). Regression analysis including a term of interaction indicated no significant relation between HGF status and benefit of treatment with abciximab ($P=0.98$) (Figure 2).

Elevated VEGF Levels Predict Increased Cardiac Risk

Patients were stratified into quartiles according to their measured VEGF levels, as follows: VEGF 1, <0.137 pg/L (n=272); VEGF 2, 0.137 to 0.289 pg/L (n=272); VEGF 3, 0.290 to 0.525 pg/L (n=272); and VEGF 4, >0.525 pg/L

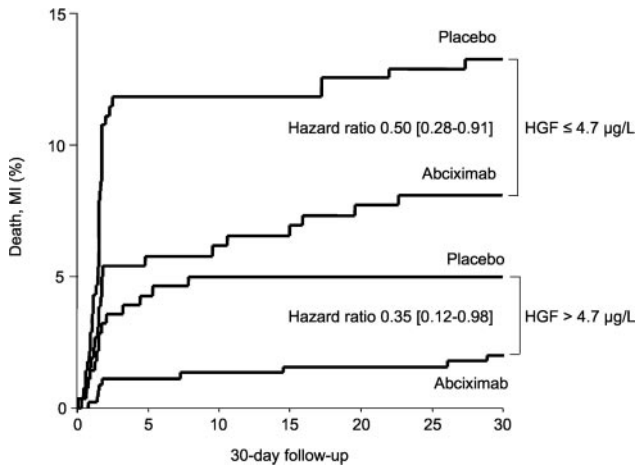


Figure 2. Adjusted hazard ratios (including 95% CIs) for treatment with abciximab according to HGF status. Benefit of treatment is defined as reduction of death or nonfatal myocardial infarction at 30-day follow-up. Hazard ratios <1.0 indicate benefit for treatment with abciximab compared with placebo. HGF serum levels did not predict the effect of treatment with abciximab.

(n=274), respectively. VEGF concentration did positively correlate with TnT levels ($R=0.43$; $P<0.001$). TnT levels were significantly higher in the third and fourth VEGF quartiles ($P<0.001$). VEGF levels also correlated with baseline CRP levels ($R=0.15$; $P<0.001$). CRP levels were significantly higher in the fourth quartile ($P=0.009$). For the initial 24-hour period, the cardiac event rates did not differ between the quartiles (Figure 3A). For the later time points (72-hour, 30-day, and 6-month follow-up), the event rate was significantly higher in the third ($P=0.001$, $P<0.001$, and $P<0.001$, respectively) and fourth quartile of VEGF serum levels ($P=0.033$, $P=0.015$, and $P=0.003$, respectively) (Figures 3A and 3B). Receiver operating characteristics curve analysis indicated a threshold level of 0.3 pg/L VEGF for maximized predictive value.

Therefore, the study population was dichotomized according to this threshold level of 0.3 pg/L, resulting in 535 patients with VEGF levels equal or above 0.3 pg/L (49.1%). Patients with high VEGF levels more frequently had ST-segment depression, transient ST-segment elevation, recurrent ischemia (21.7% versus 14.3%; $P=0.005$), CK-MB elevation (21.3% versus 8.3%; $P<0.001$), and troponin elevation (57.9% versus 18.9%; $P<0.001$) (Table 1). The unadjusted hazard ratios were 3.82 for the 72-hour follow-up (95% CI, 2.08 to 7.02; $P<0.001$), 3.11 for the 30-day follow-up (1.82 to 5.33; $P<0.001$), and 2.03 for the 6-month follow-up (1.35 to 3.06; $P<0.001$). After adjusting for the difference in baseline characteristics, VEGF levels remained predictive for patient outcome (6-month follow-up: adjusted hazard ratio 2.50 [1.52 to 4.82]; $P=0.002$).

Regression analysis, including a term of interaction, indicated a moderate relation between VEGF levels and benefit of treatment with abciximab ($P=0.046$). For patients with VEGF values ≥ 0.3 pg/L, the event rates showed a significant difference between placebo and abciximab treatment (13.0% versus 6.4%, adjusted hazard ratio 0.46 [95% CI, 0.25 to

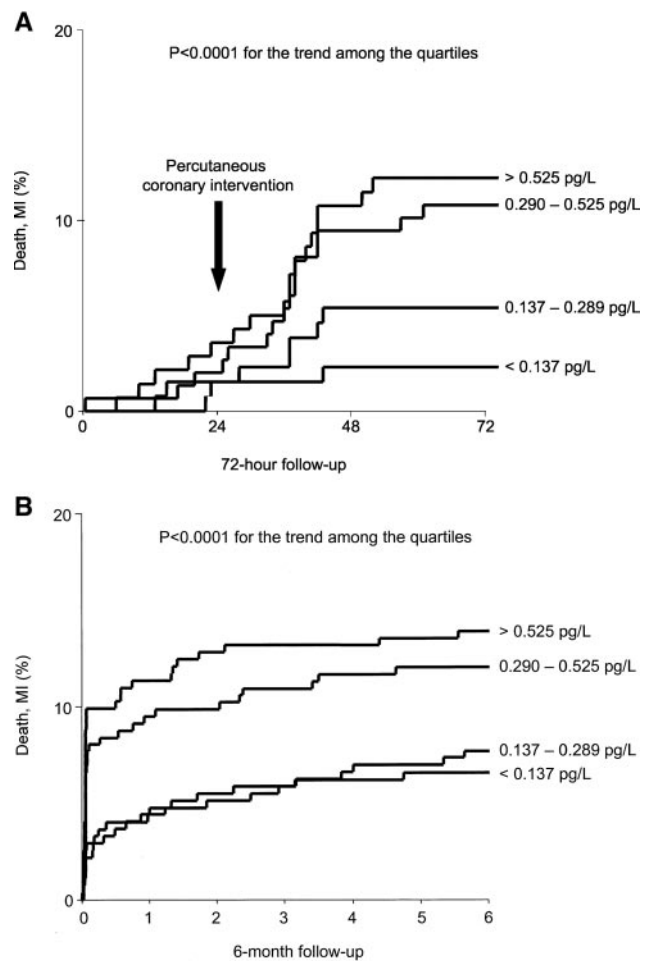


Figure 3. Association between VEGF serum levels and cardiac event rate. Patients in the upper 2 quartiles had a significantly worse outcome compared with patients in the first quartile at 72-hour follow-up (A). This early difference in cardiac risk was maintained at 6-month follow-up (B).

0.84] $P=0.007$). For patients with VEGF values <0.3 pg/L, the difference in event rates at 30-day follow-up between placebo and abciximab group was similar but did not reach statistical significance because of greater confidence intervals (4.6% versus 2.4%; adjusted hazard ratio, 0.54 [95% CI, 0.20 to 1.30]; $P=0.11$) (Figure 4).

Basic FGF Levels Are Not Related to Cardiac Risk

Patients were stratified into quartiles according to their measured basic FGF levels, as follows: bFGF 1, <5.40 $\mu\text{g/L}$ (n=271); bFGF 2, 5.40 to 9.20 $\mu\text{g/L}$ (n=271); bFGF 3, 9.21 to 15.8 $\mu\text{g/L}$ (n=271); and bFGF 4, >15.8 $\mu\text{g/L}$ (n=277), respectively. For all time points, no significant differences were observed in the event rates between the quartiles. For the 6-month follow-up, the crude event rates were as follows: first quartile, 7.3%; second quartile, 7.3% ($P=1.00$ compared with the first quartile); third quartile, 8.5% ($P=0.86$); and fourth quartile, 13.3% ($P=0.15$). Logistic regression analysis using a threshold value of 15.8 $\mu\text{g/L}$ showed no significant interaction between bFGF and cardiac risk for any selected time point.

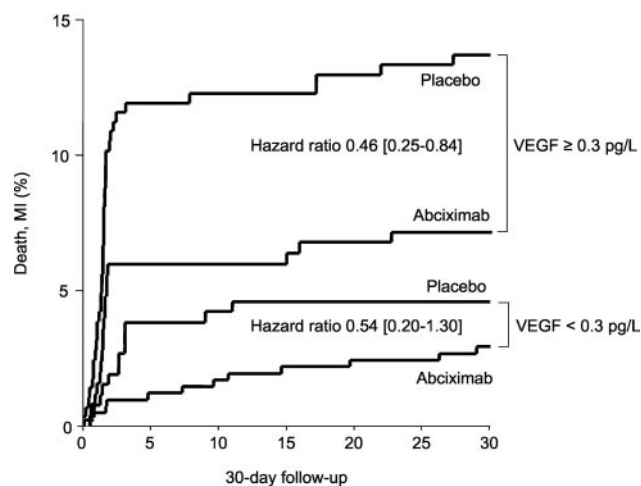


Figure 4. Adjusted hazard ratios (including 95% CIs) for treatment with abciximab according to VEGF. Benefit of treatment is defined as reduction of death or nonfatal myocardial infarction at 6-month follow-up. Hazard ratios <1.0 indicate benefit for treatment with abciximab compared with placebo. The benefit of abciximab treatment was more pronounced in patients with VEGF serum levels ≥ 0.3 pg/L.

Angiographic Features in Relation to Serum Levels of Angiogenic Growth Factors

Lesion Characteristics

In 30.2% of patients with high HGF levels, type $\geq B2$ or C lesions were documented compared with 30.1% in patients with low HGF levels ($P=0.92$). Respective values for VEGF were 30.9% versus 29.5% ($P=0.38$) and for bFGF were 30.7% versus 30.1% ($P=0.93$).

TIMI Flow

TIMI flow ≤ 1 was documented for 8.6% of patients with high HGF levels compared with 9.2% for patients with low HGF levels ($P=0.72$). In contrast, TIMI flow ≤ 1 was documented for 10.3% in patients with high VEGF levels compared with 7.6% for patients with low VEGF levels ($P=0.15$). TIMI flow at baseline was normal in 55.2% of patients with high VEGF levels versus 65.0% of patients with low VEGF levels ($P=0.008$). For bFGF, no differences with respect to TIMI flow were documented.

Thrombus

Thrombus was visible in 7.6% of patients with high HGF levels and in 5.9% of patients with low HGF levels ($P=0.35$). In patients with high VEGF levels, thrombus was visible at the time of allocation in 7.8% of patients, compared with 5.9% for patients with low VEGF levels ($P=0.28$). Respective values for bFGF were 6.5% and 7.0% ($P=0.92$).

Visible Collateral Formation

Collaterals were detectable in 22.4% of patients with high HGF levels but in only 10.5% of patients with low HGF levels ($P<0.001$). Partial or complete retrograde filling was observed in 10.0% of patients with high HGF levels versus 5.6% of patients with low HGF levels ($P<0.001$). For patients with high VEGF levels, collaterals were detectable in 17.3% of the patients, compared with 15.3% for patients with

low VEGF levels ($P=0.64$). Respective values for bFGF were 14.4% and 14.0% ($P=0.69$).

Multivariate Analysis

To delineate a potential independent prognostic significance of individual angiogenic growth factor serum levels, a multivariate analysis of the placebo group was performed including the biochemical markers TnT, CRP, HGF, and VEGF, the use of heparin, as well as baseline characteristics that revealed significant predictive value in a univariate model. Detection of collateral formation on the baseline angiogram did not predict patients' outcome. For the end points death and nonfatal myocardial infarction at 30-day and 6-month follow-up, none of the established risk factors was an independent predictor after the dichotomized biochemical markers were introduced into the model (Table 2). TnT, HGF, and VEGF remained the only independent significant predictors of patients' outcome, whereas CRP lost significance after the other biochemical markers were introduced into the model.

Discussion

The findings of the present study establish HGF serum levels as a novel independent prognostic determinant of clinical outcome in patients suffering from acute coronary syndromes. Elevated HGF serum levels are associated with a profoundly reduced incidence of death and nonfatal myocardial infarction. Most notably, in patients without troponin elevation, reduced HGF serum levels identify a subgroup of patients that suffers from significantly increased cardiac risk (adjusted hazard ratio, 8.20 [2.43 to 27.60]; $P<0.001$). In contrast, elevated VEGF serum levels are associated with worse clinical outcome in patients with acute coronary syndromes, whereas bFGF serum levels do not provide any significant information for risk stratification in these patients.

HGF, initially regarded as a growth factor specific to hepatocytes, is now well established to be a multipotent growth factor whose receptor c-met is not only expressed in the normal epithelium of almost every tissue but also in different cell types, including cardiac myocytes, endothelial

TABLE 2. Multivariate Cox Proportional-Hazards Regression Model of the Placebo Group for Death and Nonfatal Myocardial Infarction at 6-Month Follow-Up

Variable	Hazard Ratio	95% CI	P
Sex	0.85	0.65 to 1.08	0.10
Age >65 y	1.52	0.95 to 2.65	0.16
Diabetes mellitus	1.15	0.843 to 1.46	0.64
Hypercholesterolemia	0.90	0.68 to 1.13	0.59
ST-segment depression	1.33	0.82 to 1.98	0.36
Complex coronary lesions	1.15	0.73 to 2.84	0.63
Visible thrombus	1.22	0.54 to 3.61	0.92
Heparin dose	1.02	0.79 to 1.43	0.99
TnT ≥ 0.1 $\mu\text{g/L}$	1.82	1.25 to 2.59	0.03
CRP ≥ 10.0 mg/L	1.36	0.82 to 1.95	0.23
HGF ≥ 4.7 $\mu\text{g/L}$	0.30	0.16 to 0.58	<0.001
VEGF ≥ 0.3 pg/L	2.34	1.52 to 4.28	0.004

cells, and hematopoietic cells.²⁵ The general activities of HGF were found to be mitogenesis, motogenesis (enhancement of cell motility),²⁵ morphogenesis, and promotion of cell survival.²⁶ Accordingly, HGF can potentially modify several important processes contributing to the obvious beneficial effects of high HGF serum levels in patients with acute coronary syndromes. HGF has been shown to be a potent angiogenic factor, suggesting an important function during ischemic damage.²⁷ Indeed, our data showed more frequently partial or complete retrograde filling of the target vessel via collaterals in patients with elevated HGF levels. However, the presence of collaterals and subsequent retrograde filling of the target vessel was not a significant independent predictor of patients' outcome, suggesting that other mechanism than collateralization may also contribute to the protective effect of elevated HGF levels.

In this respect, several experimental studies provide evidence that HGF may also play a role for the protection from myocardial ischemia/reperfusion injury. Ono et al¹³ demonstrated the upregulation of HGF and its receptor in an experimental model of myocardial infarction, indicating that the HGF system may be part of a defense mechanism that occurs with acute myocardial ischemia. Importantly, expression of HGF also rapidly increases in other than the ischemic tissue, whereas the HGF receptor is selectively upregulated in the ischemic myocardium. Given that HGF is a strong promoter of cell survival, it is tempting to speculate that HGF exerts its cardioprotective function via cardioprotrophic effects on ischemic myocardium. Indeed, Nakamura et al¹⁵ observed that administration of HGF to rats with ischemia/reperfusion injury resulted in a significant reduction of myocardial apoptosis and infarct area and better cardiac function, whereas neutralizing antibodies to HGF resulted in profoundly increased infarct size and enhanced mortality attributable to cardiac failure.

In addition, vascular endothelial cells are a major target of HGF.¹³ HGF was experimentally shown to restore ischemia-induced vascular dysfunction to maintain endothelium-dependent regulation of coronary blood flow.¹⁷ Moreover, HGF augments the growth of hematopoietic progenitor cells.²⁸ Recent data demonstrate that a subset of hematopoietic progenitor cells importantly contributes to endothelial regeneration after endothelial denudation.²⁹ Thus, besides its potent angiogenic action, HGF may induce regeneration of the endothelial layer because of its direct antiapoptotic and cell survival promoting effects on endothelial cells. Taken together, elevated HGF serum levels may contribute to the observed improved clinical outcome in patients with acute coronary syndromes via a combination of cardioprotective functions on ischemic myocardium and vasculoprotective functions by enhancing endothelial regeneration of ruptured or eroded plaques. Thus, therapeutic interventions that enhance HGF serum levels could potentially be beneficial in patients with acute coronary syndromes.

A potential limitation for the use of HGF for risk stratification of patients with acute coronary syndromes at the time of presentation may be that serum levels of HGF are significantly elevated within minutes of heparin infusion. All patients in the CAPTURE trial were taking heparin when

serum samples were collected. It is important to notice that heparin use, dose, and activity did not differ between patients with high and low HGF serum levels (Table 1). Because heparin is standard therapy in patients with acute syndromes, blood sampling should be scheduled after the initiation of treatment.

In contrast to the favorable effects of elevated HGF serum levels, elevated VEGF serum levels were significantly associated with worse clinical outcome. Increased VEGF levels have been observed as an early response to myocardial ischemia or infarction.¹² Because VEGF is activated by the transcription factor hypoxia-induced factor-1, elevation of VEGF levels is regarded as an early adaptation of the myocardium to deprivation of blood flow.^{12,13} Indeed, in CAPTURE patients, VEGF serum levels were linked to TnT elevation, impaired TIMI flow, and clinical evidence of myocardial ischemia. Thus, VEGF serum levels measured during the acute phase of an ischemic event may actually represent a temporal marker of acutely jeopardized myocardium and, thus, provide important prognostic information. In this respect, the results of our study also demonstrate that VEGF is a powerful and independent predictor of patients' outcome after acute coronary syndromes.

In addition, recent studies suggested that activated platelets are a major source of VEGF release.²¹ Consistent with this finding, we found a more pronounced treatment effect of the potent platelet inhibitor abciximab in patients with elevated VEGF serum levels (Figure 4). Combined with previous reports demonstrating a beneficial effect of aggressive platelet inhibition by the glycoprotein IIb/IIIa inhibitor abciximab in patients with elevated troponin levels³⁰ and a significant correlation between troponin T and VEGF serum levels, these findings are consistent with the hypothesis that elevated VEGF serum levels in patients with acute coronary syndromes may predict an increased risk attributable to myocardial cell necrosis secondary to thrombus formation at the culprit lesion and distal embolization in the coronary vascular bed. Surprisingly, however, difference in the detectable thrombus formation between patients with high and low VEGF serum levels was not significant. However, it should be appreciated that angiography is not a very sensitive method for the detection of thrombus, particularly when very strict criteria are applied, as in the present study. In contrast, no interaction was found between HGF serum levels and the effect of abciximab. These findings indicate that, in contrast to elevated VEGF serum levels, elevated HGF levels do not merely reflect an increased risk of acute myocardial ischemia but rather mediate cardiovascular protective effects independent of thrombus formation at the culprit lesion.

In our study population, bFGF serum levels did not serve as a significant predictor of patients' outcome. Patients with stable coronary heart disease, enrolled in a previous study,³¹ had median bFGF serum levels of 2.4 $\mu\text{g/L}$ (95% CI, 0.2 to 13.8), whereas present patients with acute coronary syndromes enrolled in the CAPTURE trial had significantly higher bFGF serum levels with 12.1 $\mu\text{g/L}$ (95% CI, 1.2 to 72.5) ($P=0.003$). However, we did not find a significant difference in cardiac risk among patients with acute coronary syndromes when categorized according to bFGF serum lev-

els. These data suggest that, although bFGF was experimentally shown to increase collateral circulation and restore myocardial perfusion and function,^{32,33} alterations in endogenous bFGF serum levels during acute coronary syndromes do not appear to exert a significant protective effect.

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