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Role of circulating vascular endothelial growth factor and hepatocyte growth factor in patients with coronary artery disease

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Abstract Vascular endothelial growth factor (VEGF) and hepatocyte growth factor (HGF) are thought to stimulate endothelial cell proliferation and induce angiogenesis in vivo. However, the precise mechanism responsible for VEGF and HGF release in patients with coronary artery disease is still unknown. We studied serum concentrations of VEGF and HGF in 20 patients with acute myocardial infarction (AMI), 20 patients with stable angina pectoris (AP) who had reversible perfusion defects on stress myocardial scintigraphy, and 16 patients with old myocardial infarction (OMI) who had no reversible defects on stress myocardial scintigraphy. The control group consisted of 20 patients with atypical chest pain who had angiographically normal coronary arteries. Serum VEGF and HGF concentrations were measured by enzyme-linked immunosorbent assay. Both the serum VEGF and HGF concentrations in the early stage of myocardial infarction in the patients with AMI were higher than those in the patients with AP and with OMI, and control patients. The VEGF concentration in the patients with AP was higher than in the patients with OMI, whereas the HGF concentration did not differ in the patients with AP and OMI. The VEGF concentration in AMI patients who had had preinfarction angina on admission was higher than that of patients who had had no preinfarction angina, whereas the HGF concentration did not differ between the two groups of patients. These results suggest that the serum VEGF concentration may reflect myocardial ischemia to a greater degree than the serum HGF concentration.

Key words Vascular endothelial growth factor · Hepatocyte growth factor · Coronary artery disease · Myocardial ischemia

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Introduction

Vascular endothelial growth factor (VEGF) is a 38- to 46-kDa heparin-binding homodimeric glycoprotein, which is an important endothelial cell mitogen that can also enhance vascular permeability in a number of normal tissues.[1–3] The upregulation of VEGF expression in cardiac myocytes, vascular smooth muscle cells, and endothelial cells is induced by hypoxia[4–8] and myocardial ischemia.[9–12] In addition, exogenous administration of VEGF in vivo has been shown to augment collateral blood flow to the ischemic myocardium.[13,14]

Hepatocyte growth factor (HGF) is a recently characterized growth factor that has a disulfide-linked heterodimer structure and an apparent molecular weight of 80 kDa.[15–17] Its receptor has been identified as c-Met, a transmembrane tyrosine kinase proto-oncogene.[17] HGF has been shown to have numerous functions including mitogenesis,[18–20] motogenesis,[21,22] morphogenesis,[23] and angiogenesis.[24,25] Furthermore, HGF and c-Met have been implicated in capillary endothelial cell regeneration in the ischemically injured heart.[26]

Recently, circulating concentrations of VEGF and HGF were found to have increased after acute myocardial infarction (AMI) in humans.[27–32] However, no studies have compared the changes in these growth factors in a variety of coronary syndromes. Therefore, the present study was designed to determine serum VEGF and HGF concentrations in patients with AMI, angina pectoris (AP), or old myocardial infarction (OMI).

Subjects and methods

Patients

We studied 20 patients with first AMI (mean age 64 years) who were hospitalized at our coronary care unit within 3 h after the onset of symptoms, 20 patients with AP (mean age

69 years) who had angiographic evidence of ≧90% coronary stenosis and had reversible perfusion defects on stress myocardial scintigraphy, and 16 patients with OMI (mean age 64 years) who had angiographic evidence of ≧90% coronary stenosis in one vessel and no reversible perfusion defects on stress myocardial scintigraphy. The control group consisted of 20 patients (mean age 65 years) with atypical chest pain who had angiographically normal coronary arteries and no evidence of myocardial ischemia.

AMI was diagnosed based on typical chest pain lasting >30 min with >0.2 mV of ST-T segment elevation in at least two contiguous leads on a standard 12-lead electrocardiogram and an increase in the creatine kinase-MB activity that was at least twice the upper normal limit. AP was defined as typical anginal chest pain lasting between 10 and 30 min with ≧1 mm transient ST-segment depression or elevation 0.08 s after the J point, and no episodes of angina at rest or increasing angina. The serum creating kinase and creatine kinase-MB activities had to be less than twice the upper limit of normal. Patients with OMI were enrolled in the study at least 6 months after the occurrence of AMI.

Adenosine triphosphate (ATP) stress thallium-201 myocardial scintigraphy

ATP was infused for 5 min at a rate of 0.16 mg/kg per min via an antecubital vein. Three minutes after the start of ATP infusion, 74 MBq of thallium-201 was injected as a bolus at another venous site. Single photon emission computed tomographs were obtained 5 min and 4 h after the first thallium injection using a large field-of-view rotating gamma camera equipped with a low-energy, high-resolution collimator interfaced to a computer (Toshiba NEW GMS-550U, Toshiba Medical, Tokyo, Japan). Four hours later, 37 MBq of thallium-201 was reinjected and the imaging was repeated. Tomographic slices were displayed in all three standard cardiac planes to assess myocardial perfusion in each vascular territory. The presence or absence of redistribution was determined visually from the 4-h images. Thallium-201 images were interpreted by two nuclear cardiologists who did not know the results of the coronary angiography.

Coronary angiography

All patients with AP and OMI underwent coronary angiography within 2 weeks of ATP stress thallium-201 myocardial scintigraphy. Coronary angiography was performed using the standard Judkins technique. Patients with coronary artery stenoses that were ≧90% of the vessel diameter in one or more epicardial vessels were enrolled in this study. The results were interpreted by two senior angiographers who had no knowledge of the results of the thallium imaging.

Blood samples

Venous blood samples for the measurement of VEGF and HGF were obtained immediately before the administration of heparin at the time of admission in patients with AMI. In the AP, OMI, and control groups, fasting venous blood samples were obtained at 0700 h. Clotted cellular elements were removed from the blood samples by centrifugation at room temperature (2000 × *g*, 10 min). The serum samples were stored at −70°C until the time of assay.

Biochemical analysis

Serum VEGF and HGF were concentrations measured by enzyme-linked immunosorbent assay (VEGF, Immuno-Biological Laboratories, Gunma, Japan; HGF, Otsuka Assay Laboratories, Tokyo, Japan). The sensitivity of the VEGF and HGF kits was 15.6 pg/ml and 0.010 ng/ml, respectively.

Statistical analysis

All results are expressed as the mean ± standard deviation. The differences in ratios were compared using the chi-squared test. Differences among the four groups were evaluated by one-factor analysis of variance, and if it was significant, Fisher's Protected least significance difference test was performed. Relationships were analyzed using the Pearson correlation coefficient. A value for $P < 0.05$ was considered statistically significant.

Results

Patient characteristics

Table 1 summarizes the patient characteristics in each study group. The groups were comparable with respect to gender, age, incidence of hypertension and diabetes mellitus, and total cholesterol and triglyceride concentrations. The smoking rate was higher in the AMI group than in the control group. The HDL-cholesterol concentration was higher in the control group than in the other groups, and lower in the AMI group than in the AP group. Ejection fraction on left ventriculography was higher in the AP and control groups than in the AMI and OMI groups.

Antiplatelet drugs including aspirin, ticlopidine, and trapidil were administered to 1 of 20 control patients, 12 of 20 AP patients, and all of OMI patients. In all AMI patients, antiplatelet drugs were not administered on admission. However, in patients with AP, there were no differences between those receiving and not receiving antiplatelet drugs with respect to the concentrations of serum VEGF and HGF. Antihyperlipidemic drugs such as statin were administered to 2 of 20 control patients, 5 of 20 AP patients, 3 of 16 OMI patients, and 4 of 20 patients with AMI on admission, the ratios of which did not differ.

The VEGF and HGF concentrations did not differ between the patients with and without hypertension or diabetes mellitus either overall or in any group.

Table 1. Patient demographics and clinical characteristics

	Group			
	AMI	AP	OMI	Control
n	20	20	16	20
Male/female	15/5	12/8	11/5	10/10
Age (years)	63.7 ± 12.5	66.9 ± 11.0	63.6 ± 9.4	64.8 ± 11.0
		*		
Smokers (*n*)	10 (50.0%)	5 (25.0%)	5 (31.3%)	3 (15.0%)
Hypertension (*n*)	11 (55.0%)	11 (55.0%)	5 (31.3%)	5 (25.0%)
Diabetes mellitus (*n*)	5 (25.0%)	6 (30.0%)	4 (25.0%)	5 (25.0%)
T-Cholesterol (mg/dl)	194.8 ± 35.1	205.0 ± 37.0	203.5 ± 39.8	201.7 ± 34.6
Triglycerides (mg/dl)	124.6 ± 59.6	151.5 ± 61.5	119.9 ± 43.1	131.4 ± 53.6
		**		
		*		**
HDL-Cholesterol (mg/dl)	40.5 ± 9.1	46.8 ± 6.1	40.9 ± 12.4	52.5 ± 9.2
			*	
		*		
		**	**	**
LVEF (%)	55.4 ± 15.2	68.6 ± 7.3	55.4 ± 7.4	66.3 ± 8.7

Data are reported as the mean ± standard deviation
AMI, acute myocardial infarction; AP, angina pectoris; OMI, old myocardial infarction; T-, total; HDL-, high-density lipoprotein; LVEF, left ventricular ejection fraction
* $P < 0.05$; ** $P < 0.01$

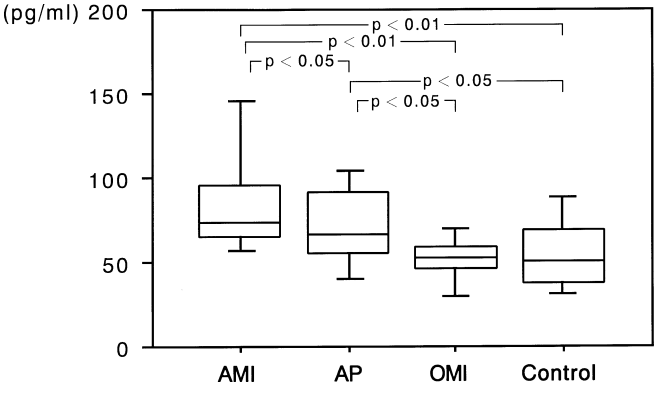

Fig. 1. Serum vascular endothelial growth factor concentrations in the control group and patients with acute myocardial infarction (*AMI*), angina pectoris (*AP*), and old myocardial infarction (*OMI*)

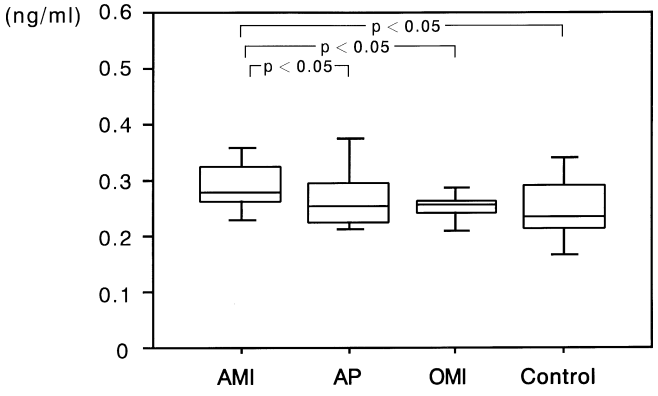

Fig. 2. Serum hepatocyte growth factor concentrations in the control group and patients with acute myocardial infarction (*AMI*), angina pectoris (*AP*), and old myocardial infarction (*OMI*)

Serum VEGF concentrations (Fig. 1)

The serum VEGF concentration was higher at the time of admission in patients with AMI than in the AP, OMI, and control groups (AMI group, 89.6 ± 44.0 pg/ml; AP group, 74.5 ± 29.0 pg/ml; OMI group, 54.7 ± 21.2 pg/ml; control group, 56.5 ± 26.8 pg/ml). The VEGF concentration was higher in the patients with AP than in the OMI and control groups. The VEGF concentration in the patients with OMI did not differ from that of control patients.

Serum HGF concentrations (Fig. 2)

The serum HGF concentration was higher at the time of admission in the patients with AMI than in the AP, OMI, and control groups (AMI group, 0.30 ± 0.06 ng/ml; AP group, 0.27 ± 0.07 ng/ml; OMI group, 0.25 ± 0.04 ng/ml; control group, 0.25 ± 0.06 ng/ml). The HGF concentration did not differ between the AP, OMI, and control groups.

Correlation between serum VEGF and HGF concentrations (Fig. 3)

The VEGF concentration did not correlate with the HGF concentration in the AMI ($r = 0.021$), AP ($r = 0.102$), OMI ($r = 0.093$), or control ($r = 0.230$) groups.

Correlation between serum growth factor concentrations and the angiographic findings (Table 2)

Patients in the coronary artery disease subgroups (AMI, AP, and OMI groups) were divided into four groups based on the angiographically determined development of collaterals

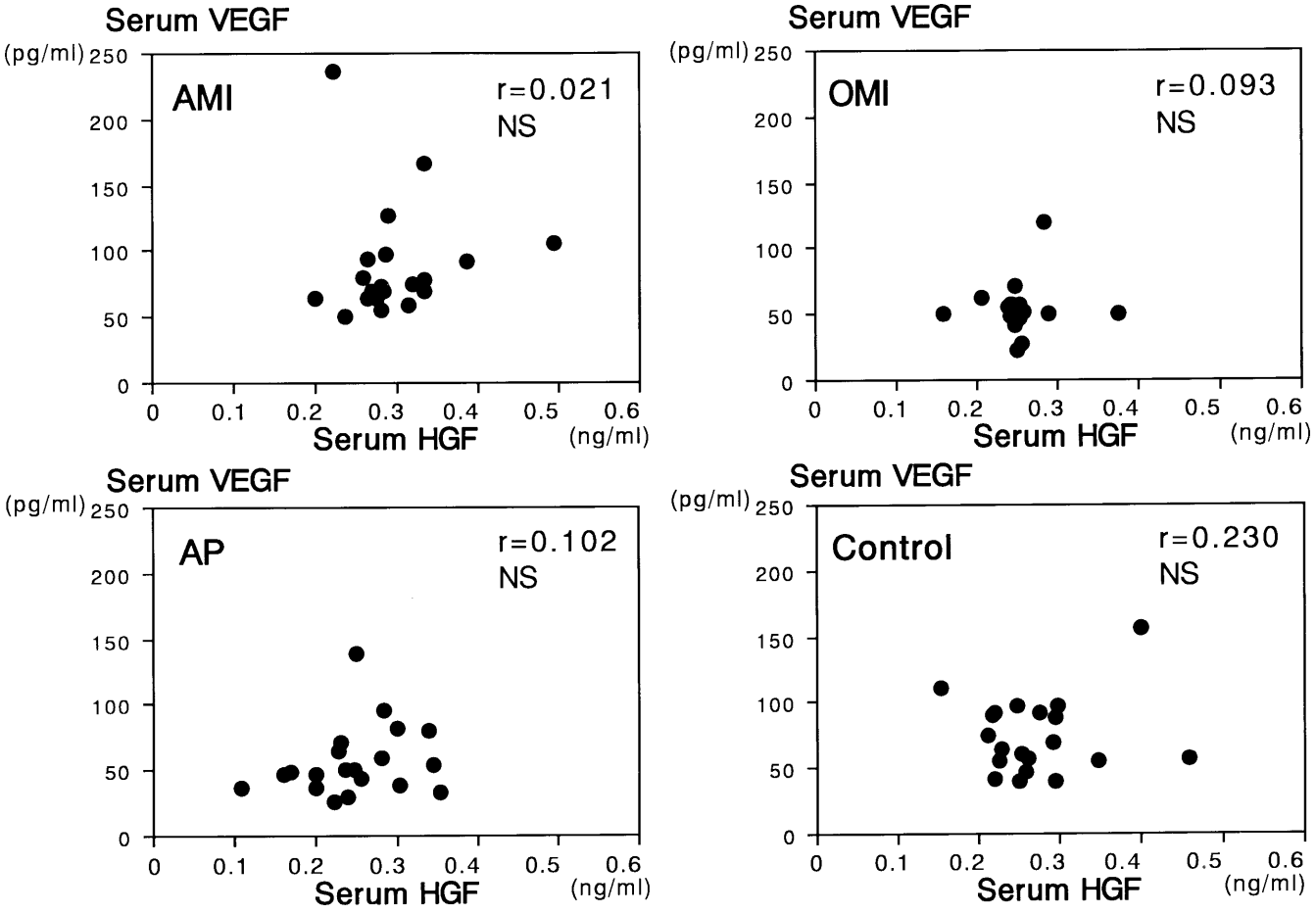

Fig. 3. Correlation between the serum vascular endothelial growth factor (*VEGF*) and serum hepatocyte growth factor (*HGF*) concentrations. *NS*, not significant

Table 2. Correlations between serum growth factor concentrations and angiographic findings

	Spearman's *r*	*P* value
AMI		
Collateral score/VEGF	0.034	0.8898
Collateral score/HGF	0.245	0.3029
AP		
Collateral score/VEGF	−0.161	0.5038
Collateral score/HGF	0.066	0.7857
OMI		
Collateral score/VEGF	0.109	0.6924
Collateral score/HGF	0.361	0.1730

VEGF, vascular endothelial growth factor; HGF, hepatocyte growth factor; other abbreviations as in Table 1

using the system designed by Rentrop et al.[33] (grades 0–4). The serum VEGF and HGF concentrations did not correlate in any group with the degree of collateral development.

Serum VEGF and HGF concentrations of AMI with or without preinfarction angina in the AMI group

The VEGF concentration in AMI patients who had had preinfarction angina on admission (8 patients) was higher than that of patients who had had no preinfarction angina (12 patients) (113.2 ± 59.7 vs 74.0 ± 20.1 pg/ml, $P < 0.05$), whereas the HGF concentration did not differ between the patients with and without preinfarction angina (0.310 ± 0.09 vs 0.286 ± 0.04 ng/ml).

Discussion

In this study, the serum VEGF and HGF concentrations were both higher at the time of admission in the patients with AMI than in the AP, OMI, or control patients. These results might be due to increased expression of VEGF and HGF induced by the myocardial ischemia.[9–12,26] The degree of myocardial ischemia might explain the differences in serum VEGF and HGF concentrations between the AMI and AP patients. In a model of myocardial infarction in rats, an initial rapid rise in the mRNA expression of VEGF and its receptors (*flk-1*, *flt-1*) was observed throughout the entire heart.[11] Furthermore, the plasma concentrations of HGF also increases within 1 h of reperfusion after 1 h of myocardial ischemia.[26] In the present study, the circulating concentrations of VEGF and HGF were elevated within 3 h after the onset of AMI, suggesting that the rapid induction

of VEGF and HGF mRNA followed by synthesis of the proteins might occur in response to acute myocardial ischemia in humans. Several clinical reports have demonstrated that the serum HGF concentration increases at the time of admission in patients with AMI,[29–32] which is in keeping with our results. In contrast, the changes in the serum concentration of VEGF in patients with AMI is controversial. Seko et al.[27] demonstrated that the serum VEGF concentration before reperfusion is markedly increased in patients with AMI. However, Tamura et al.[28] and Kranz et al.[29] reported that the serum VEGF concentration in patients with AMI does not increase immediately after admission. In their studies, no detailed characteristics of the subjects were presented, such as sex and mean age, which makes analysis of the different results between their studies and ours difficult. For example, it could not be denied that the control groups in their studies had silent myocardial ischemia. Furthermore, the serum VEGF concentration in the patients with AMI in the present study (89.6 pg/ml) comparable to that in the study by Kranz et al. (105 pg/ml) is less than half of those in the studies by Seko et al. (252.4 pg/ml) and Tamura et al. (213 pg/ml). This discrepancy might have resulted from differences in duration from the onset because the patients with AMI in the study by Tamura et al. were within 24 h of the onset longer than 3 h in our criteria. The use of different biochemical analysis kits might also have contributed to this discrepancy.

Previous study showed that the serum HGF concentration increased within 3 h after the onset in patients with AMI, which is compatible with our results. However, whether the serum VEGF concentration increases in the very early stage in AMI is still unknown. In this study, the serum VEGF concentration also increased within 3 h after the onset in AMI and was higher in the AMI patients than in the other groups. Thus, the measurement of VEGF might be useful for the early diagnosis of AMI if a more rapid assay for VEGF is developed.

In the present study, the serum VEGF concentration in the patients with AP was greater than in those with OMI and control patients. In contrast, the HGF concentration in the patients with AP was comparable to that in patients with OMI and control patients. These results suggest that the serum VEGF concentration reflects myocardial ischemia better than the serum HGF concentration. In other words, VEGF might be upregulated mainly by ischemia, while HGF might be upregulated by other stimuli. This hypothesis is supported by our finding that the VEGF concentration did not correlate with the HGF concentration in any group. Also, the VEGF concentration in the AMI patients who had had preinfarction angina on admission was higher than that of patients who had not experienced preinfarction angina. However, Kranz et al. have shown that the serum VEGF concentration was not increased in patients with unstable angina.[29] In their study, no detailed characteristics of the control group, such as angiographic findings, were presented, which makes analysis of the different results between the two studies difficult. In addition, differences in biochemical analysis (they used an immunoradiometric assay) might have caused the difference in the results.

HGF activator, a serum-derived serine protease, has been identified as an activator of the single-chain form of HGF.[34] The HGF activator precursor is converted to the active form by thrombin.[35] Recent studies have shown that increases in the circulating HGF concentration can be induced by arterial thrombus formation.[36] Furthermore, we were able to detect an apparent thrombus by coronary angiography in 2 of 20 AMI patients, who demonstrated a higher concentration of serum HGF (0.39 and 0.32 ng/ml) than the mean value of that in AMI patients (0.30 ng/ml). Therefore, a coronary thrombus might induce an increase in the serum HGF concentration in the early stage of AMI through thrombin generation. Furthermore, the serum HGF concentration could reflect differences in the degree of thrombus deposition between AP and AMI.

Previous studies have also demonstrated that VEGF is often extensively expressed in human coronary arteries narrowed by atherosclerotic plaque.[37] The serum HGF concentration is increased in patients with coronary atherosclerosis or severe retinal arteriosclerosis,[38] suggesting that VEGF and HGF expression might be associated with the severity of atherosclerosis. However, in the present study, no differences in the VEGF and HGF concentrations were found between the OMI and control groups, which suggests that the VEGF and HGF concentrations might not directly reflect the severity of coronary atherosclerosis. Another previous study has demonstrated that the serum HGF concentration increased in hypertensive patients and decreased in diabetic patients without hypertension.[39] However, in the present study, the serum HGF concentration did not differ between the patients with and without hypertension or diabetes either overall or in any one group.

VEGF and HGF also have potent angiogenic effects in animal models; specifically, they improve the collateral blood flow in the ischemic myocardium[13,14] and ischemic hindlimb muscle.[40–42] In the present study, neither the serum VEGF concentration nor the HGF concentration correlated with the degree of collateral development in any of the groups with coronary artery disease. This result suggests that the circulating VEGF and HGF concentrations may not directly reflect the effects of the local VEGF/HGF system on collateral development. This study included a small number of patients, therefore the ability to generalize this correlation might be limited.

Experimental studies have demonstrated that HGF produced by transfection with a human HGF vector can exert stimulatory autocrine and paracrine effects on endothelial cell growth.[43] In a rat model, HGF and c-Met have been implicated in capillary endothelial cell regeneration in the ischemically injured heart.[26] Furthermore, increased local vascular HGF production can prevent endothelial injury.[44] These findings suggest that one role of increased circulating HGF in AMI might be the repair of injured endothelial cells.

Our study has several limitations. First, we could not perform serial measurements of serum VEGF and HGF concentrations in the acute phase of myocardial infarction

because heparin administration reduces the serum VEGF[28] and increases the serum HGF concentration.[45] Second, we measured only the serum concentrations of VEGF and HGF and did not evaluate the local concentrations of VEGF and HGF in the ischemic regions. Further studies based on blood sampling from the coronary sinus and aorta are needed to determine the main site of VEGF/HGF production in patients with coronary artery disease.

References

1. Senger DR, Galli SJ, Dvorak AM, Harvey VS, Dvorak HF (1983) Tumor cells secrete a vascular permeability factor that promotes accumulation of ascites fluid. Science 219:983–985
2. Connolly DT, Heuvelman DM, Nelson R, Olander JV, Eppley BL, Delfino NR, Siegal RM (1989) Tumor vascular permeability factor stimulates endothelial cell growth and angiogenesis. J Clin Invest 84:1470–1478
3. Ferrara N, Houck KK, Jakeman LL, Leung DW (1992) Molecular and biological properties of the vascular endothelial growth family of proteins. Endocrinol Rev 13:18–32
4. Shweiki D, Itin DA, Soffer D, Keshet E (1992) Vascular endothelial growth factor induced by hypoxia may mediate hypoxia-initiated angiogenesis. Nature 359:843–845
5. Ladoux A, Frelin C (1993) Hypoxia is a strong inducer of vascular endothelial growth factor mRNA expression in the heart. Biochem Biophys Res Commun 195:1005–1010
6. Brogi E, Wu T, Namiki A, Isner JM (1994) Indirect angiogenic cytokines upregulate VEGF and bFGF gene expression in vascular smooth muscle cells, whereas hypoxia upregulates VEGF expression only. Circulation 90:649–652
7. Levy AP, Levy NS, Loscalzo J, Calderone A, Takahashi N, Yeo KT, Koren G, Colucci WS, Goldberg MA (1995) Regulation of vascular endothelial growth factor in cardiac myocytes. Circ Res 76:758–766
8. Namiki A, Brogi E, Kearney M, Kim EA, Wu T, Couffinhal T, Varticovski L, Isner JM (1995) Hypoxia induces vascular endothelial growth factor in cultured human endothelial cells. J Biol Chem 270:31189–31195
9. Banai S, Shweiki D, Pinson A, Chandra M, Lazarovici G, Keshet E (1994) Upregulation of vascular endothelial growth factor expression induced by myocardial ischemia: implications for coronary angiogenesis. Cardiovasc Res 28:1176–1179
10. Hashimoto E, Ogita T, Nakaoka T, Matsuoka R, Takao A, Kira Y (1994) Rapid induction of vascular endothelial growth factor expression by transient ischemia in rat heart. Am J Physiol 267: H1948–H1954
11. Li J, Brown LF, Hibberd MG, Grossman JD, Morgan JP, Simons M (1996) VEGF, flk-1, and flt-1 expression in a rat myocardial infarction model of angiogenesis. Am J Physiol 270:H1803–H1811
12. Sellke FW, Wang SY, Stamler A, Lopez JJ, Li J, Simons M (1996) Enhanced microvascular relaxations to VEGF and bFGF in chronically ischemic porcine myocardium. Am J Physiol 271: H713–H720
13. Banai S, Jaklitsch MT, Shou M, Lazarous DF, Scheinowitz M, Biro S, Epstein SE, Unger EF (1994) Angiogenic-induced enhancement of collateral blood flow to ischemic myocardium by vascular endothelial growth factor in dog. Circulation 89:2183–2189
14. Asahara T, Bauters C, Zheng LP, Takeshita S, Bunting S, Ferrara N, Symes JF, Isner JM (1995) Synergistic effect of vascular endothelial growth factor and basic fibroblast growth factor on angiogenesis in vivo. Circulation 92:II-365–II-371
15. Nakamura T, Nawa K, Ichihara A (1984) Partial purification and characterization of hepatocyte growth factor from serum of hepatectomized rats. Biochem Biophys Res Commun 122:1450–1459
16. Nakamura T, Nishizawa T, Hagiya M, Seki T, Shimonishi A, Sugimura A, Tashiro K, Shimizu S (1989) Modular cloning and expression of human hepatocyte growth factor. Nature 342: 440–443
17. Gherardi E, Stroker M (1991) Hepatocyte growth factor-scatter factor: mitogen, motogen and met. Cancer Cells 3:227–232
18. Rubin JS, Chan AM-L, Bottaro DP, Burgess WH, Taylor WG, Cech AC, Hirschfield DW, Wong J, Miki T, Finch PW, Aaronaon SA (1991) A broad-spectrum human lung fibroblast-derived mitogen is a variant of hepatocyte growth factor. Proc Natl Acad Sci USA 88:415–419
19. Strain AJ, Ismail T, Tsubouchi H, Arakaki N, Hishida T, Kitamura N, Daikuhara Y, McMaster P (1991) Native and recombinat human hepatocyte growth factors are highly potent promoters of DNA synthesis in both human and rat hepatocytes. J Clin Invest 87:1853–1857
20. Takahashi M, Ota S, Shimada T, Hamada E, Kawabe T, Okudaira T, Matsumura M, Kaneko N, Terano A, Nakamura T, Omata M (1995) Hepatocyte growth factor is the most potent endogenous stimulant of rabbit gastric epithelial cell proliferation and migration in primary culture. J Clin Invest 95:1994–2003
21. Stoker M, Gherardi E, Perryman M, Gray J (1987) Scatter factor is a fibroblast-derived modulator of epithelial cell mobility. Nature 327:239–242
22. Grey A-M, Schor AM, Rushton G, Ellis I, Schor SL (1989) Purification of the migration stimulating factor produced by fetal and breast cancer patient fibroblasts. Proc Natl Acad Sci USA 86:2438–2442
23. Montesano R, Matsumoto K, Nakamura T, Orci L (1991) Identification of a fibroblast-derived epithelial morphogen as hepatocyte growth factor. Cell 67:901–908
24. Bussolino F, Di Renzo MF, Ziche M, Bocchietto E, Olivero M, Naldini L, Gaudino G, Tamagnone L, Coffer A, Comoglio PM (1992) Hepatocyte growth factor is a potent angiogenic factor which stimulates endothelial cell motility and growth. J Cell Biol 119:629–641
25. Grant DS, Kleinman HK, Goldberg ID, Bhargava MM, Nickoloff BJ, Kinsella JL, Polverini P, Rosen EM (1993) Scatter factor induces blood vessel formation in vivo. Proc Natl Acad Sci USA 90:1927–1941
26. Ono K, Matsumori A, Shioi T, Furukawa U, Sasayama S (1997) Enhanced expression of hepatocyte growth factor/c-Met by myocardial ischemia and reperfusion in a rat model. Circulation 95:2552–2558
27. Seko Y, Iami Y, Suzuki S, Kamijukkouku S, Hayasaki K, Sakomura Y, Tobe K, Kadowaki T, Maekawa H, Takahashi N, Yazaki Y (1997) Serum levels of vascular endothelial growth factor in patients with acute myocardial infarction undergoing reperfusion therapy. Clin Sci 92:453–454
28. Tamura K, Nakajima H, Rakue H, Ssame A, Naito Y, Nagai Y, Ibukiyama C (1999) Elevated circulating levels of basic fibroblast growth factor and vascular endothelial growth factor in patients with acute myocardial infarction. Jpn Circ J 63:357–361
29. Kranz A, Rau C, Kochs M, Waltenberger J (2000) Elevation of vascular endothelial growth factor-A serum levels following acute myocardial infarction. Evidence for its origin and functional significance. J Mol Cell Cardiol 32:65–72
30. Matsumori A, Furukawa Y, Hashimoto T, Ono K, Shioi T, Okada M, Iwasaki A, Nishio R, Sasayama S (1996) Increased circulating hepatocyte growth factor in the early stage of acute myocardial infarction. Biochem Biophys Res Commun 221:391–395
31. Sato T, Yoshinouchi T, Sakamoto T, Fujieda H, Murao S, Sato H, Kobayashi H, Ohe T (1997) Hepatocyte growth factor (HGF): a new biochemical marker for acute myocardial infarction. Heart Vessels 12:241–246
32. Sato T, Yoshinouchi T, Sugimoto T, Sakamoto T, Fujieda H, Murao S, Sato H, Ohe T (1999) Prognostic value of serum hepatocyte growth factor in patients with acute coronary syndromes. Jpn Circ J 63:583–588
33. Rentrop KP, Cohen M, Blanke H, Phillips RA (1985) Changes in collateral channel filling immediately after controlled coronary artery occlusion by an angioplasty balloon in human subjects. J Am Coll Cardiol 5:587–592
34. Miyazawa K, Shimomura T, Kitamura A, Kondo J, Morimoto Y, Kitamura N (1993) Molecular cloning and sequence analysis of the cDNA for a human serine protease responsible for activation of hepatocyte growth factor. Structural similarity of the protease precursor to blood coagulation factor XII. J Biol Chem 268:10024–10028

35. Shimomura T, Kondo J, Ochiai M, Naka D, Miyazawa K, Morimoto Y, Kitamura N (1993) Activation of the zymogen of hepatocyte growth factor activator by thrombin. J Biol Chem 268:22927–22932
36. Matsumori A, Ono K, Furukawa Y, Okada M, Sasayama S (1998) Circulating hepatocyte growth factor as an early marker of arterial thrombus formation. Jpn Circ J 62:311–313
37. Couffinhal T, Kearney M, Witzenbichler B, Chen D, Murohara T, Losordo DW, Symes J, Isner JM (1997) Vascular endothelial growth/vascular permeability factor (VEGF/VPF) in normal and atherosclerotic human arteries. Am J Pathol 150:1673–1685
38. Nishimura M, Ushiyama M, Nanba A, Ohtsuka K, Yoshimura M (1998) Vascular endothelium-related factors and atherosclerosis/arteriosclerosis: serum hepatocyte growth factor as a possible indicator of vascular lesions. Jpn J Clin Pathol 46:671–677
39. Morishita R, Moriguchi A, Higashi J, Ogihara T (1999) Hepatocyte growth factor (HGF) as a potential index of severity of hypertension. Hypertens Res 22:161–167
40. Rakue H, Nakajima H, Katoh T, Usui M, Amemiya T, Miyagi M, Hara T, Tamura K, Sasame A, Naito Y, Nagai Y, Ibukiyama C (1998) Low-dose basic fibroblast growth factor and vascular endothelial growth factor for angiogenesis in canine acute hindlimb insuffiency. Jpn Circ J 62:933–939
41. Grant DS, Kleinmann HK, Goldberg ID, Bhargava MM, Nickoloff BJ, Kinsella JL, Polverini P, Rosen EM (1993) Scatter factor induces blood vessel formation in vivo. Proc Natl Acad Sci USA 90:1937–1941
42. Van Belle E, Witzenbichler B, Chen D, Silver M, Chang L, Schwall R, Isner JM (1998) Potentiated angiogenic effect of scatter factor/hepatocyte growth factor via induction of vascular endothelial growth factor: the case for paracrine amplification of angiogenesis. Circulation 97:381–390
43. Hayashi S, Morishita R, Higashi J, Aoki M, Moriguchi A, Kida I, Yoshiki S, Matsumoto K, Nakamura T, Kaneda Y, Ogihara T (1996) Autocrine-paracrine effects of overexpression of hepatocyte growth factor gene on growth of endothelial cells. Biochem Biophys Res Commun 220:539–545
44. Morishita R, Higaki J, Hayashi SI, Yo Y, Aoki M, Nakamura S, Moriguchi A, Matsushita H, Matsumoto K, Nakamura T, Ogihara T (1997) Role of hepatocyte growth factor in endothelial regulation: prevention of high D-glucose-induced endothelial cell death by prostaglandins and phosphodiesterase type 3 inhibitor. Diabetologia 40:1053–1061
45. Okada M, Matsumori A, Ono K, Miyamoto T, Takahashi M, Sasayama S (1999) Hepatocyte growth factor is a major mediator in heparin-induced angiogenesis. Biochem Biophys Res Commun 255:80–87