PROGNOSTIC SIGNIFICANCE OF SERUM VASCULAR ENDOTHELIAL GROWTH FACTOR AND HEPATOCYTE GROWTH FACTOR IN PATIENTS WITH ACUTE CORONARY SYNDROME

(Thesis)

Submitted for partial fulfillment of the M.D. Degree in Cardiology

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Acknowledgement

I would like to express my deepest thanks to Dr. Khaled Sorour, M.D., Professor of Cardiology, Faculty of Medicine, Cairo University, for his kind and generous attitude and his great help and supervision throughout this work.

Also, I would like to express my deepest appreciation to Dr. Yasser Baghdady, M.D., Assistant Professor of Cardiology, Faculty of Medicine, Cairo University, for his great help, advice, assistance and valuable ideas he had offered and for the time he spent with me to make this work valid.

My thanks to Dr. Heba Raslan, M.D., Lecturer of Clinical Pathology, Cairo University for the great effort she made during the laboratory studies, also her valuable advice and suggestions.

I would like to express my deepest gratitude to Dr. Basma Elgamal, M.D., Assistant Professor of Clinical Pathology and Hematologic Oncology, Cairo University, for her great help, scientific support, wise direction, valuable advice and suggestions. And I am really indebted for her patience and kindness.

Last, but not least, my sincere appreciation is for my family, who supported me a great deal and endured so many hard times until I succeeded by God's Will to accomplish this work.

Abstract

Coronary artery disease is one of the most prevalent diseases worldwide. In acute coronary syndrome, up regulation of angiogenic growth factors as a part of the compensatory mechanisms aims to restore the blood flow to the ischemic tissue. Among these angiogenic factors, are hepatocyte growth factor (HGF) and vascular endothelial growth factor (VEGF). However, it is unknown whether or not; the level of these angiogenic growth factors is related to clinical outcome. We measured VEGF and HGF levels in ٥٠ patients with a mean age of ٥٩.٣±٥.٧٨ years, as well as ١٠ age and sex matched healthy individuals as a control group. Then the patients were divided into two groups; group A with high VEGF level, and group B with high HGF level. Angiogenic evaluation of these patients was performed. Then the patients were followed for a period of six months for any ischemic manifestations, such as myocardial infarction, cerebral stroke and death. At the end of six months, all the patients performed stress test to detect residual ischemia. We found significant elevation of HGF and VEGF in patients with acute coronary syndrome as compared to the controls ($p < ٠.٠٠١$), and the total event rate in the group of patients with high HGF was significantly lower than that in the group of patients with high VEGF ($p < ٠.٠٠١$).

Conclusion: Angiogenic growth factors; VEGF and HGF could be independent predictors of patient prognosis in acute coronary syndrome, whereas, VEGF elevation is correlated to adverse outcome, HGF elevation was associated with a favorable prognosis.

Key words: Acute coronary syndrome, angiogenesis, VEGF, HGF

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List of Abbreviations

ACH	Acetyl choline
ACS	Acute coronary syndrome
ADP	Adenosine diphosphate
AML	Acute myeloid leukemia
CAD	Coronary artery disease
CAPS	Coronary artery bypass surgery
CFR	Coronary flow reserve
CFU-GEMM	Colony forming unit-granulocyte, erythrocyte, monocyte, megakaryocyte
c-GMP	Cyclic guanylate monophosphate
CHD	Coronary heart disease
CHF	Congestive heart failure
CRP	C-reactive protein
DCs	Dendritic cells
DNA	Deoxy ribo nucleic acid
ECs	Endothelial cells
EDHF	Endothelial derived hyperpolarizing factor
EDRF	Endothelial derived relaxing factor
EGF	Epidermal growth factor
EPC	Endothelial progenitor cell
ET	Endothelin
FGF	Fibroblast growth factor
HCC	Hepato cellular carcinoma
HGF	Hepatocyte growth factor
HIFs	Hypoxia inducible transcrption factors
HSC	Hematopoietic stem cell
K-ATP	Adenosine triphosphate sensitive potassium channel
kb	Kilo base
KDa	Kilo Dalton
LDL	Low density lipoprotein
LH	Leutinizing hormone
L-NMMA	N-monmethyl L-arginine
LP(a)	Lipoprotein a
LV	Left ventricle
MDS	Myelodysplastic syndrome
MI	Myocardial infarction
MMP	Metalloproteinases
mRNA	Messenger ribo nucleic acid
MVO٢	Myocardial ventilation oxygen consumption
NFK-B	Nuclear factor kappa beta
NO	Nitric oxide
NOS	Nitric oxide synthetase
OA	Osteoarthritis
PAF	Platelet activating factor
PCI	Percutaneous coronary intervention
PDGF	Platelet derived growth factor
PDR	Proliferative diabetic retinopathy
PET	Positron emission tomography
PIGF	Platelet induced growth factor
RA	Rheumatoid arthritis
RV	Right ventricle
SMCs	Smooth muscle cells
SPECT	Single photon emission computed tomography
TNF	Tumor necrosis factor
UA	Unstable angina
VEGF	Vascular endothelial growth factor

Introduction

In the recent years, biochemical markers have contributed to better understanding of underlying pathophysiological processes in patients with acute coronary syndrome; markers of myocardial necrosis have become a valuable tool for risk stratification and optimizing treatment strategies **(de Winter et al., ١٩٩٥)**. During consecutive repair phase after the onset of acute coronary syndrome, compensatory processes are initiated, including the formation of collateral circulation as well as endothelial regeneration of the ruptured or eroded plaques **(Heeschen et al., ٢٠٠٠)**.

A variety of potent angiogenic growth factors have been identified to stimulate neovascularization of ischemic tissue **(Isner and Asahara, ١٩٩٩)**. Among these, vascular endothelial growth factor and hepatocyte growth factor have been experimentally shown to improve tissue perfusion when administered exogenously into ischemic tissue **(Van Belle et al., ١٩٩٨)**. In addition, both vascular endothelial growth factor and hepatocyte growth factor have been shown to be up-regulated endogenously after myocardial infarction **(Lee et al., ٢٠٠٠)**.

However, it is still unknown how serum levels of these angiogenic factors relate to clinical outcome in patients with acute coronary syndrome **(Heeshen et al., ٢٠٠٣)**.

Hepatocyte growth factor, 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 on normal

epithelium of almost every tissue, but also in different cell types including cardiac myocytes, endothelial cell and hematopoietic cells **(Matsumoto and Nakamura, ١٩٩٦)**. The general activities of hepatocyte growth factor were found to be mitogenesis, motogenesis **(Matsumoto and Nakamura, ١٩٩٦)**, morphogenesis and promotion of cell survival **(Zarnegar and Michalopoulos ١٩٩٥)**.

Vascular endothelial growth factor is a highly specific mitogen for vascular endothelial cells. The most striking property of vascular endothelial growth factor is its specificity, it is mitogenic for capillary endothelial cells, but not for other types of cells **(Matsumoto and Claesson-Welsh, ٢٠٠١)**.

Aim of Work

The aim of this work is to estimate the serum level of vascular endothelial growth factor and hepatocyte growth factor in patients with acute coronary syndrome and to investigate whether or not these angiogenic factors have a prognostic relevance in such patients.

Myocardial Ischemia and Regulation of Coronary Blood Flow

Myocardial ischemia is a condition in which oxygen deprivation to the heart muscle is accompanied by inadequate removal of metabolites because of reduced blood flow or perfusion. In contrast, mere oxygen deprivation (hypoxia or anoxia) without reduction in the clearance of metabolites occurs in cyanotic congenital heart disease, cor pulmonale, severe anemia, asphyxiation, and carbon monoxide poisoning. Patients with these problems do not exhibit ischemic symptoms **(Braunwald, ١٩٧١)**.

Frequency:

- **In the US:** The prevalence of myocardial ischemia is difficult to ascertain since the actual numbers of ischemic episodes and patients with myocardial ischemia would have to include those with silent ischemia. Silent ischemia is undoubtedly common, but its precise prevalence is unknown. Atherosclerotic coronary heart disease (CHD) causes approximately ٥٠٠,٠٠٠ deaths in the United States each year, ie, about ١ in ٥ deaths overall. Roughly ٦.٣ million Americans are believed to experience angina. An estimated ٣٥٠,٠٠٠ new cases of angina occur every year. More than ١٢ million Americans had a history of MI and/or angina pectoris in the year ٢٠٠٠. About every ٢٩ seconds, an American has a coronary

event, and about every minute someone dies from one **(Shekher, ٢٠٠٥)**.

- **Internationally:** International incidence, especially in the developed countries, echoes that observed in the United States **(Gheorghiade and Bonow, ١٩٩٨)**.

Mortality/Morbidity: In the United States, approximately ١٤ million persons have ischemic heart disease and its various complications. CHF, as a result of ischemic cardiomyopathy, has become the most common discharge diagnosis in US hospitals.Approximately ١.٥ million Americans have acute MI annually, ٥٠٠,٠٠٠ of who die. CAD is the single most common cause of death in the United States, with a rate of almost ١ death per minute. More than half of those who die suddenly from CAD have no previous symptoms. In the United States, almost ٥٠٠,٠٠٠ patients undergo PCIs, and another ٤٠٠,٠٠٠ individuals undergo coronary artery bypass surgery (CABS) per year as a result of myocardial ischemia and/or MI **(Shekher, ٢٠٠٥)**.

Survivors of MI exhibit a poorer prognosis as well. They have a ١.٥- to ١٥-times higher risk of mortality and morbidity than the rest of the population without prior MI and are at higher risk for subsequent MI, as well as for fatal and near-fatal arrhythmias as a result of myocardial ischemia. Within a year of MI, ٢٥% of men and ٣٨% of women die. Within ٦ years, ١٨% of men and ٣٤% of women have a second MI, ٧% of men and ٦% of women experience sudden death, ٢٢% of men and

٤٦% of women are disabled with CHF, and ٨% of men and ١١% of women have a stroke **(Shekher, ٢٠٠٥)**.

Race: Significant racial variations exist in the incidence, prevalence, presentations, and response to therapy for CAD. African Americans appear to have higher morbidity and mortality rates, even when corrected for educational and socioeconomic status **(Singh, ١٩٩٣)**. Indian Asians have ٢- to ٣-times higher incidence of CAD than whites in the United States. They have greater incidence of hypoalphalipoproteinemia and high lipoprotein (a) (Lp(a)) levels **(Raitakari et al., ١٩٩٩)**. Persons of Mediterranean origin have a lower incidence of CAD and myocardial ischemia. The relative importance of genetics versus environment (especially diet) has yet to be completely elucidated in this population **(Singh, ١٩٩٣)**.

Sex: The incidence of ischemic heart disease is equal in men and women, with the onset in women typically delayed by about ١٠ years. Premenopausal women are generally protected from manifestations of ischemic heart disease because of the protective effects of estrogen, but the presence of diabetes eliminates the protection associated with female sex. The mortality rates associated with ischemic heart disease are similar for both men and women, although they are slightly higher in women.

Numerous studies have reported that women tend to present later than men when presenting with acute MI or ischemic episodes, are less often subjected to invasive strategies, and tend to benefit less from these invasive strategies. Earlier studies tended to show women having a greater overall mortality rate during invasive strategies, but this has been contradicted in more recent studies **(Singh, ١٩٩٣)**.

Age: Age is one of the strongest independent risk factors for development of CAD and all its presenting manifestations. Elderly persons still experience higher mortality and morbidity rates from CAD than younger people. Complications of multiple therapeutic interventions tend to be higher **(Yusuf et al., ٢٠٠٠)**.

History: Ischemic heart disease and myocardial ischemia are manifested in a broad spectrum of clinical syndromes. The spectrum of presentation includes symptoms and signs consistent with the following conditions **(Krone, ١٩٩٨)**:

- Asymptomatic state (subclinical phase)
- Stable angina pectoris
- Unstable angina (acute coronary syndrome)
- Acute MI
- Chronic ischemic cardiomyopathy
- CHF
- Sudden cardiac arrest

For any of the clinical syndromes described above, the patient may report one or more of the following signs and symptoms **(Knatterud et al., ١٩٩٤)**:

- Chest discomfort
- Shortness of breath
- Fatigue and reduced exertional capacity due to ischemia-mediated cardiac dysfunction
- Palpitations and dizziness from arrhythmias that occur as a result of myocardial ischemia
- Leg swelling and weight gain from heart failure

- Symptoms related to the risk factors for CAD
- Silent myocardial ischemia

Pathophysiology:

During ischemia, an imbalance occurs between myocardial oxygen supply and demand. Ischemia may manifest as (١) anginal discomfort, (٢) ST-segment deviation on ECG, (٣) reduced uptake of thallium ٢٠١ or technetium ٩٩ in myocardial perfusion images, or (٤) regional or global impairment of ventricular function **(Miller et al., ١٩٩٤)**. Myocardial ischemia can occur as a result of increased myocardial oxygen demand, reduced myocardial oxygen supply, or both. In the presence of coronary obstruction, an increase of myocardial oxygen requirements caused by exercise, tachycardia, or emotion leads to a transitory imbalance. This condition is frequently termed *demand ischemia* and is responsible for most episodes of chronic stable angina. In other situations, the imbalance is caused by acute reduction of oxygen supply secondary to increased coronary vascular tone (i.e., coronary vasospasm) or by marked reduction or cessation of coronary flow as a result of platelet aggregates or thrombi. This condition, termed supply ischemia, is responsible for myocardial infarction (MI) and most episodes of unstable angina (UA). In many circumstances, ischemia results from both an increase in oxygen demand and a reduction in supply **(Anwaruddin et al., ٢٠٠٥)**.

The heart is an aerobic organ and therefore relies almost exclusively on the oxidation of substrates for generation of energy. It can develop only a small oxygen debt and still have enough energy to

function normally. Thus, in a steady state, determination of the rate of myocardial oxygen consumption (i.e., rate of myocardial ventilation oxygen consumption [MVO_2]) provides an accurate measure of its total metabolism. That the total metabolism of the arrested. The small fraction of MVO_2 in the noncontracting heart is required for those physiologic processes not directly associated with contraction. Increases in the frequency of depolarization of the noncontracting heart are accompanied by only small increases in myocardial oxygen consumption **(Braunwald, ٢٠٠٠)**.

Determinants of myocardial oxygen consumption

- Heart rate
- Contractility
- Systolic wall tension
- Shortening against a load (Fenn effect)
- Maintenance of cell viability in basal state
- Depolarization
- Activation
- Maintenance of active state
- Direct metabolic effect of catecholamines
- Fatty acid uptake

Heart rate: Myocardial oxygen consumption is linearly related to heart rate, i.e., the faster the ventricular rate, the greater the myocardial oxygen consumption **(Takaoka et al., ١٩٩٣)**.