Prognostic Value of Circulating Levels of Endothelin-1 in Patients After Acute Myocardial Infarction Undergoing Primary Coronary Angioplasty*

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Background: The link between increased circulating level of endothelin (ET)-1 and adverse clinical outcomes after acute myocardial infarction (AMI) has been established. Current studies demonstrate that reperfusion therapy by either thrombolysis or primary percutaneous coronary intervention (PCI) can salvage myocardium, improving survival of AMI patients. However, whether reperfusion therapy by primary PCI can prevent the adverse effect of ET-1 on clinical outcomes in patients after AMI remains unclear. Therefore, this study examined the predictive value of circulating ET-1 levels on 30-day outcomes in ST-segment elevated AMI treated with primary PCI.

Methods and results: We conducted a prospective cohort study of 186 consecutive patients with ST-segment elevated AMI of onset < 12 h who underwent primary PCI. Blood samples for plasma concentration of ET-1 were collected in the catheterization laboratory following vascular puncture. Patients were classified into a high group (group 1, ET-1 level > 0.632 pg/mL, n = 93) and a low group (group 2, ET-1 level < 0.632 pg/mL, n = 93) according to the median value of ET-1 after AMI. Univariate analysis demonstrated that the 30-day composite major adverse clinical outcomes (MACO) [advanced Killip score ≥ 3], severe congestive heart failure (CHF) [New York Heart Association functional class ≥ 4], and 30-day mortality were strongly associated with high ET-1 level (p < 0.0001), unsuccessful reperfusion (final Thrombolysis in Myocardial Infarction flow ≤ 2; p < 0.0001), low left ventricular ejection fraction (< 50%; p = 0.0002), multivessel disease (p = 0.005), and female gender (p = 0.007). Multiple stepwise logistic regression analysis demonstrated that only high ET-1 level (p < 0.0001) and unsuccessful reperfusion (p < 0.0001) were independent predictors of 30-day MACO. Additionally, high ET-1 level (p = 0.0021) along with unsuccessful reperfusion (p = 0.008) and severe CHF (p < 0.0001) were significant independent predictors of increased 30-day mortality.

Conclusions: A high circulating level of ET-1 is a strong independent predictor of 30-day MACO after ST-segment elevated AMI treated with primary PCI. (CHEST 2005; 127:1491–1497)

Key words: acute myocardial infarction; endothelial-1; primary coronary angioplasty

Abbreviations: AMI = acute myocardial infarction; CHF = congestive heart failure; DM = diabetes mellitus; ET = endothelin; LVEF = left ventricular ejection fraction; MACO = major adverse clinical outcomes; MB = myocardial blush; PCI = percutaneous coronary intervention; TIMI = Thrombolysis in Myocardial Infarction

Acute myocardial infarction (AMI) is accompanied by metabolic and neurohormonal changes that may relate to the severity of illness and clinical outcomes. Rapidly advancing secretion of neurohormones by neuroendocrine systems is a physiologic response to myocardial damage in patients with AMI.

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in order to maintain a hemodynamic homeostasis. However, the overreactive responses of these hormones, eg, vasoconstriction and tachycardia, resulted in catecholamines,\(^3\) which might be harmful to the patients. Considerable clinical evidence shows that increased neuroendoctrine activity is detrimental to patients with clinical setting of AMI.\(^{1,3,4}\)

Endothelin (ET)-1, a 21 amino acid residue peptide made by endothelial cells, is the most potent endogenous vasoconstrictor yet identified.\(^5,6\) It has been reported that circulating ET-1 increases in the early hours after AMI.\(^7,8\) After AMI, higher and more prolonged elevations of this vasoconstrictor have been discovered in patients with a complicated clinical course\(^7\) and left ventricular failure,\(^7,9,10\) and have consequently been closely associated with increased mortality.\(^10,11\) However, the precise pathophysiologic effects of ET-1 in AMI patients remain uncertain. Animal model has demonstrated that ET-1 may contribute to microvasculature dysfunction due to its potent vasoconstrictive property, thus having an adverse effects in AMI by restricting myocardial blood flow following reperfusion.\(^8\) However, whether ET-1 also has a concordant effect on microvasculature dysfunction in human beings with AMI, when undergoing primary percutaneous coronary intervention (PCI) as in the animal model remains unreported.

Current data have demonstrated that reperfusion therapy by either thrombolysis or primary PCI improves left ventricular function and patient survival.\(^12,13\) The purpose of this study was to examine the predictive value of circulating ET-1 on 30-day outcomes in ST-segment elevation AMI treated with primary PCI.

**For editorial comment see page 1474**

**Materials and Methods**

*Study Population and Inclusion Criteria*

In our hospital, all patients with AMI were considered eligible for primary PCI. For the purpose of this study, the circulating level of ET-1 of all patients undergoing primary PCI was prospectively measured. Blood samples were drawn after vascular puncture prior to coronary angiography in the cardiac catheterization room. To exclude variables that could influence the serum levels of high-sensitivity C-reactive protein, patients were excluded with a recent surgical history or trauma within the previous 2 months, renal insufficiency (creatinine > 1.5 mg/dL), malignancy, acute or chronic inflammatory disease at study entry or recent infection, as well as febrile disorders. Patients were also excluded if fever (body temperature > 37.5°C) was observed in the emergency department. Between November 2002 and December 2003, we prospectively investigated and recruited 228 consecutive patients of any age who presented with AMI and undergoing primary PCI in our hospital. Seventeen of the 203 patients (8.4%) were subsequently excluded due to fever (n = 3), infection (n = 3), malignancy (n = 2), liver cirrhosis (n = 1), or renal function impairment (n = 8). Therefore, the remaining 186 patients constituted the study population. Patients were classified into one of two groups (the high or low ET-1 groups) based on the extent of ET-1 level. The median value of ET-1 was used to classify patients into these two groups.

Thirty subjects matched for age, gender, hypertension, diabetes mellitus (DM), current smoking, and hypercholesterolemia served as risk factor control subjects. Twenty age- and gender-matched healthy volunteers were also studied. Informed consent was obtained from all study subjects, and the study protocol was approved by the Institutional Review Committee on Human Research of our institution.

**Procedure and Protocol**

A transradial artery approach using a 6F arterial sheath was routinely applied for AMI in our hospital unless results of the Allen test were positive in both hands. A 6F Kimny Miniradi (Boston Scientific, Scimed; Maple Grove, MN) was used for diagnosis and primary PCI.

Tirofiban therapy (loading dose, 20 μg/kg of body weight) was administered to patients on presentation in the emergency department, followed by a maintenance infusion of 0.15 μg/kg/min for 18 to 24 h at the beginning of this study. However, tirofiban therapy was subsequently withheld because this therapy was found not to provide an additional benefit to AMI patients who underwent primary PCI.\(^{14}\) Therefore, only 36 patients received tirofiban therapy in this study.

From May 2002, a guard wire device (PercuSurge GuardWire; Medtronic; Minneapolis, MN) was utilized in consecutive patients presenting with AMI (< 12 h in duration) in our hospital. The enrollment and exclusion criteria, and procedural method have been described previously in detail.\(^{15}\)

Clopidogrel (300 mg loading dose after stenting, then 75 mg/d) was administered for at least 4 weeks to patients who underwent primary stenting. Aspirin, 100 mg/d po, was administered to each patient indefinitely.

**Blood Sampling, Laboratory Investigations, and Assays**

Blood samples were obtained once in healthy volunteers from a healthy clinic examination and risk-control subjects in the outpatient department. Biochemical measurements were determined by standard laboratory methods.

The plasma level of ET-1 was measured using a standard enzyme-linked immunosorbent assay and a commercial kit (R&D Systems; Minneapolis, MN). The method for the enzyme-linked immunosorbent assay was described in detail in our recent report.\(^{16}\) The assay was sufficiently sensitive to detect < 1.0 pg/mL of ET-1. The cross-reactivity of ET-2, ET-3, and big ET in this assay were 45%, 14%, and < 1%, respectively, according to the manufacturer of the assay kits. Intraindividual variability of ET-1 levels was assessed in study patients, risk-control subjects, and healthy subjects. The mean intra-assay coefficients of variance were 6.10% (patients), 5.8% (risk control), and 4.7% (normal control), respectively.

**Definitions and Data Collection**

AMI was defined by the presence of typical chest pain for > 30 min with ST-segment elevation > 1 mm in two consecutive precordial or inferior leads. Unsuccessful reperfusion was designated if the infarct-related artery did not reach normal blood flow.
after PCI. Failed reperfusion of microvasculature was defined as post-PCI myocardial blush (MB) grade ≤ 1.17 Reperfusion time was defined as from onset of chest pain to the first balloon inflation. Detailed in-hospital and follow-up data were obtained, including age, sex, coronary risk factors, Killip score on admission, severity of congestive heart failure (CHF), preinfarction angina, body temperature on admission, WBC count, creatinine level, angiographic findings, and number of diseased vessels. These data were collected prospectively and entered into a computerized database.

**End Points and Statistical Analysis**

The primary end point of the study was the 30-day composite occurrence of major adverse clinical outcomes (MACO), including advanced Killip score (≥ 3), severe CHF (defined as New York Heart Association functional class 4), or 30-day death.

Data were expressed as mean ± SD. Categorical variables were compared using χ² test or Fisher exact test. Continuous variables among three groups were compared using one-way analysis of variance for parametric data and Kruskal-Wallis test for nonparametric data. Wilcoxon rank-sum test with Bonferroni correction was used to improve the normality of nonparametric data for statistical analysis. Statistical analysis was performed using statistical software (SAS for Windows Version 8.2; SAS Institute; Cary, NC). A probability value < 0.05 was considered statistically significant.

**Results**

**Baseline Characteristics of Study Patients, Angina Patients, and Healthy Control Subjects**

There were no significant differences among the three groups with regard to age and gender (Table 1). There were also no significant differences between study patients and risk control subjects in terms of coronary artery disease risk factors. However, the circulating level of ET-1 was substantially higher in patients than in risk control and normal control subjects.

**Comparison of Baseline Characteristics, Clinical Findings and Outcomes, and Angiographic Results between High and Low ET-1 Groups**

Patients with an ET-1 level < 0.632 pg/mL were classified into the low ET-1 group (group 2) and those with an ET-1 level ≥ 0.632 pg/mL were placed in the high ET-1 group (group 1) [Table 2]. The mean value of ET-1 was significantly higher in group 1 than in group 2 patients (p < 0.0001). The two groups were similar with respect to cardiovascular risk factors, previous stroke, and previous myocardial infarction, or anterior wall infarction. In addition, there were no significant differences in terms of stent implantation, utilization of PercuSurge device, or adjunctive tirofiban therapy between group 1 and group 2 patients. However, group 1 patients were older with more prevalence of female gender than group 2 patients. Furthermore, group 1 patients had significantly higher incidences of advanced Killip score and severe CHF than group 2 patients. Moreover, the duration of blood sampling times and reperfusion times was markedly longer in group 1 than in group 2.

Angiographic findings demonstrated no distinctive differences in pre-PCI Thrombolysis in Myocardial Infarction (TIMI) flow grades, post-PCI TIMI-3 flow, or MB grade ≥ 2. However, multivessel disease was significantly higher in group 1 than in group 2 patients. Additionally, left ventricular angiography demonstrated that the high ET-1 group patients had distinctively lower left ventricular ejection fraction (LVEF) than the low ET-1 group patients. Two group 1 patients with severe CHF had acute in-stent thrombus requiring repeated PCI. However, one of the group 1 patients with preexisting distal stent edge dissection had...
subacute in-stent thrombus requiring stenting. The 30-day mortality rate was substantially higher in group 1 than in group 2 patients.

**Independent Predictors of Raised Circulating Level of ET-1**

Multiple stepwise logistic regression analysis of the baseline characteristics on Table 2 demonstrated that only female gender, advanced Killip score, reperfusion time (≥ 240 min), and lower LVEF were independent predictors of elevated circulating level of ET-1 (≥ 0.632 pg/mL) [Table 3].

### Table 3—Multiple Stepwise Logistic Regression Analysis of Predictors Related to Elevated ET-1 (≥ 0.632 pg/mL)*

<table>
<thead>
<tr>
<th>Variables</th>
<th>Odds Ratio</th>
<th>95% Confidence Interval</th>
<th>p Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Female</td>
<td>3.62</td>
<td>1.38–9.47</td>
<td>0.009</td>
</tr>
<tr>
<td>Advanced Killip score*</td>
<td>2.42</td>
<td>1.05–5.56</td>
<td>0.037</td>
</tr>
<tr>
<td>Duration of AMI (≥ 240 min)†</td>
<td>2.44</td>
<td>1.25–4.74</td>
<td>0.009</td>
</tr>
<tr>
<td>LVEF, % (≥ 50%)</td>
<td>3.09</td>
<td>1.54–6.16</td>
<td>0.001</td>
</tr>
</tbody>
</table>

*Defined as Killip score ≥ 3.
†Duration of AMI was defined as chest pain onset to blood sampling time.

**Correlates of 30-Day MACO**

Univariate analysis of factors associated with 30-day MACO is shown in Table 4. Unsuccessful reperfusion and high ET-1 level were the most significant factors among these variables. Lower LVEF was the second-most-significant factor among these variables. Female gender and age ≥ 70 years were significantly related to increased 30-day MACO. A strong correlation between 30-day MACO and DM, anterior wall infarction, and multivessel disease was also present.

**Independent Correlates of 30-Day Mortality and Composite End Points**

By multiple stepwise logistic regression analysis (Table 5), only unsuccessful reperfusion, high ET-1 level, and severe CHF were significant independent predictors of increased 30-day mortality. Furthermore, the only variables independently related to 30-day MACO were unsuccessful reperfusion and high ET-1 level (Table 6).

**Discussion**

The present study, in which the circulating level of ET-1 was examined in patients with AMI undergoing
First, patients with high circulating ET-1 level had a 30-day mortality rate > 5.7 times higher than that of patients with low circulating ET-1 level. Second, the present study showed a strong independent association between increasing ET-1 and 30-day mortality. Third, the results of this study confirmed previous observations relating elevated ET-1 to 30-day mortality. These in turn lead to infarction expansion and left ventricular dysfunction, increased ischemic zone, which in turn leads to increased ET-1 secretion in order to maintain the hemodynamic status. Therefore, we suggest that high circulating ET-1 level is a poor prognostic biomarker in patients with AMI complicated by cardiogenic shock. Therefore, our finding raises issues regarding the relationship between the ischemic area and the circulating ET-1 level.

Another important finding in this investigation was that patients with AMI complicated by advanced Killip score (≥ 3) had remarkably higher ET-1 levels than those with AMI of Killip score 1 and 2. Furthermore, advanced Killip score was an independent predictor of elevated circulating level of ET-1. There could be several reasons for these clinical observations. First, a situation of advanced Killip score usually causes a slow coronary blood flow globally. Second, globally slow coronary blood flow results in global ischemia in these patients. Third, endothelial cells are more susceptible to global ischemia, which in turn leads to increased ET-1 secretion in order to maintain the hemodynamic status. However, this potent vasoconstrictor increases myocardial oxygen demand by an increase in afterload and further reduces coronary blood flow and oxygen supply by vasoconstriction of the coronary bed. These in turn lead to infarction expansion and left ventricular dysfunction, increased ischemic zone, and worsening heart failure, finally yielding a vicious cycle leading to refractory shock and death. This could explain why our patients with AMI complicated by cardiogenic shock strongly associated with 30-day mortality had significantly higher ET-1 levels. Therefore, we suggest that high circulating level of ET-1 is a poor prognostic biomarker in patients after AMI.

Another important finding in the present study was that the ET-1 level was more substantially increased in patients with subsequently severe CHF and low left ventricular function than in patients without them. Furthermore, lower LVEF was strongly associated with raised circulating level of ET-1. In addition to these findings, the study also revealed that ET-1 along with unsuccessful reperfusion was a significantly independent predictor of increased 30-day MACO. Moreover, the most important finding in

### Table 4—Univariate Analysis of Baseline Characteristics, Laboratory Findings, and Angiographic Results in Prediction of 30-d Major Adverse Clinical Outcomes

<table>
<thead>
<tr>
<th>Variables</th>
<th>Odds Ratio</th>
<th>95% Confidence Interval</th>
<th>p Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age (≥ 70 yr vs &lt; 70 yr)</td>
<td>2.08</td>
<td>1.04–4.13</td>
<td>0.037</td>
</tr>
<tr>
<td>Female vs male gender</td>
<td>2.99</td>
<td>1.35–6.62</td>
<td>0.007</td>
</tr>
<tr>
<td>DM vs non-DM</td>
<td>2.01</td>
<td>1.04–3.90</td>
<td>0.037</td>
</tr>
<tr>
<td>Hypertension vs non-hypertension</td>
<td>0.99</td>
<td>0.52–1.87</td>
<td>0.969</td>
</tr>
<tr>
<td>With vs without hypercholesterol</td>
<td>0.70</td>
<td>0.37–1.34</td>
<td>0.278</td>
</tr>
<tr>
<td>Smoking vs nonsmoking</td>
<td>0.81</td>
<td>0.43–1.54</td>
<td>0.522</td>
</tr>
<tr>
<td>Reperfusion time (&gt; 240 min vs ≤ 240 min)</td>
<td>1.72</td>
<td>0.88–3.37</td>
<td>0.114</td>
</tr>
<tr>
<td>Anterior vs non-anterior wall infarction</td>
<td>2.0</td>
<td>1.02–3.85</td>
<td>0.043</td>
</tr>
<tr>
<td>Multivessel vs single-vessel disease</td>
<td>2.63</td>
<td>1.34–5.13</td>
<td>0.005</td>
</tr>
<tr>
<td>With vs without PercuSurge use</td>
<td>0.57</td>
<td>0.29–1.12</td>
<td>0.100</td>
</tr>
<tr>
<td>With vs without tirofiban therapy</td>
<td>1.68</td>
<td>0.54–2.98</td>
<td>0.287</td>
</tr>
<tr>
<td>LVEF, % (≥ 50 vs &lt; 50)</td>
<td>0.28</td>
<td>0.14–0.55</td>
<td>0.0002</td>
</tr>
<tr>
<td>Final TIMI flow (2 vs 3)</td>
<td>7.69</td>
<td>2.86–20.0</td>
<td>&lt; 0.0001</td>
</tr>
<tr>
<td>ET-1 level (≥ 0.632 pg/mL vs &lt; 0.632 pg/mL)</td>
<td>5.09</td>
<td>2.45–10.59</td>
<td>&lt; 0.0001</td>
</tr>
</tbody>
</table>

### Table 5—Multiple Stepwise Logistic Regression Analysis of Baseline Characteristics Laboratory Findings, and Angiographic Results in Prediction of 30-d Mortality

<table>
<thead>
<tr>
<th>Variables</th>
<th>Odds Ratio</th>
<th>95% Confidence Interval</th>
<th>p Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>High ET-1 level (≥ 0.632 pg/mL)</td>
<td>6.79</td>
<td>1.34–34.42</td>
<td>0.0021</td>
</tr>
<tr>
<td>Unsuccessful reperfusion</td>
<td>7.14</td>
<td>1.69–33.30</td>
<td>0.008</td>
</tr>
<tr>
<td>Severe CHF</td>
<td>14.91</td>
<td>4.32–51.44</td>
<td>&lt; 0.0001</td>
</tr>
</tbody>
</table>

Table 6—Multiple Stepwise Logistic Regression Analysis of Baseline Characteristics Laboratory Findings, and Angiographic Results in Prediction of 30-d Major Adverse Clinical Outcomes

<table>
<thead>
<tr>
<th>Variables</th>
<th>Odds Ratio</th>
<th>95% Confidence Interval</th>
<th>p Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>High ET-1 level (≥ 0.632 pg/mL)</td>
<td>6.75</td>
<td>2.92–15.60</td>
<td>&lt; 0.0001</td>
</tr>
<tr>
<td>Unsuccessful reperfusion</td>
<td>12.50</td>
<td>3.70–33.33</td>
<td>&lt; 0.0001</td>
</tr>
</tbody>
</table>
the present study was that the high ET-1 level along with unsuccessful reperfusion and severe CHF, which have been recognized to be poor prognostic risk factors following AMI, were significant independent predictors of increased 30-day mortality. Therefore, the results of our study, besides consistently with previous studies, explored that the impact of high ET-1 level on MACO and 30-day mortality could not be abolished by the use of the primary PCI. This finding is clinically important because it encourages the use of this powerful parameter for the risk stratification of AMI patients.

An age of ≥70 years and female gender were significantly related to increased circulating level of ET-1 after AMI in the present study, but the causes for the significantly higher level of circulating ET-1 in these patients are not fully understood. However, the contributive factor may be that the preexisting endothelial dysfunction, which is more vulnerable to ischemic injury, is present in the older patients. Furthermore, perhaps fundamental differences in the biology and pathophysiology of AMI between men and women may be another reason that concerns this clinical observation. Therefore, it is not surprising that our results based on the clinical observation could, at least in part, explain why women and the aged have worse prognostic outcomes after AMI.

Previous studies have shown that elevated circulating ET-1 is observed in the early hours of myocardial infarction. In the present study, we found that patients with longer time-to-treatment interval had markedly higher circulating ET-1 level than those who had shorter time-to-treatment interval. Furthermore, the duration of AMI was independently associated with elevated circulating level of ET-1. Our findings corroborate with previous results further extending the concept that a concomitant increased circulating ET-1 level is proportional to ischemic duration. This finding could partially explain why previous studies have shown that the timing of reperfusion is strongly associated with 30-day clinical outcomes in patients with AMI following reperfusion therapy.

There were several limitations on this study. First, without serial measurement of circulating ET-1 levels, we did not provide information regarding to how early the circulating ET-1 elevated and how long the elevation of ET-1 persisted in our patients. Second, experimental study has demonstrated that ET-1 may contribute to microvasculature dysfunction due to its potent vasoconstrictive property, thus having an adverse effect in AMI by reducing myocardial blood flow after reperfusion. However, no correlation was found between elevated circulating ET-1 level and unsuccessful reperfusion or failed reperfusion of microvasculature. This may be probable due to either the small sample size of unsuccessful reperusions in our study substantially distorting the true meaning of the results, or due to different reperfusion methods in humans vs animal studies. The effects of circulating ET-1 on long-term clinical outcomes were not part of the designed protocol; therefore, these relationships did not extend beyond the 30-day clinical outcomes.

In conclusion, higher circulating ET-1 level on admission is strongly associated with the increased 30-day MACO independent of the utilization of primary PCI. This relationship may explain the higher mortality rates observed among AMI patients with an increased circulating ET-1 level and help to clarify the important role of ET-1 levels for risk stratification of patients with AMI.

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