Plasma intermedin levels in patients with acute myocardial infarction

Zhengbing Lv[a,1], Kai Wu[b,1], Xiaoping Chen[a,*], Xin Zhang[a], Biying Hong[a]

[a] Department of Cardiology, West China Hospital, Sichuan University, Chengdu, Sichuan 610041, China
[b] Department of Cardiology, 903 Hospital, Jiangyou, Sichuan 621700, China

ARTICLE INFO

ABSTRACT

It has been shown that adrenomedullin (ADM) may function as a cardiovascular-regulatory peptide in humans. Intermedin (IMD) is a newly discovered peptide related to ADM and has a greater range of biological effects on the cardiovascular in animal experiments. The purpose of the study was to investigate the pathophysiological role of IMD in patients with acute myocardial infarction (AMI). The present study included twenty patients with acute ST-segment elevation myocardial infarction (STEMI), thirty-three with stable coronary heart disease (SCHD), and eighteen healthy controls. Plasma levels of IMD, malonaldehyde (MDA), and superoxide dismutase (SOD) and cardiac biomarkers were determined at one, two, four and seven days following AMI. Plasma IMD levels were significantly increased on day 1 in AMI patients when compared with SCHD subjects ($P = 0.014$), and reached a peak of 181.88 ± 9.47 pg/ml at 96 h. Plasma IMD concentrations were correlated with MDA and SOD. Furthermore, patients with severe lesions in their coronary arteries tended to have higher plasma IMD levels ($P < 0.05$) in AMI patients. A significant increase in plasma IMD following AMI may be associated with oxidative stress, and could be used as a marker to reflect the severity of the coronary stenosis.

© 2013 Elsevier Inc. All rights reserved.

1. Introduction

Cardiovascular diseases are a leading cause of mortality and morbidity according to estimates by the World Health Organization (WHO) [13]; however, the mechanisms of these diseases remain unclear. Vascular endothelial cell damage, insulin resistance and atherosclerosis induced by adipose infiltration, inflammation and oxidative stress play critical roles in the pathogenesis of coronary heart disease and its complications [14,16,23].

Intermedin (IMD), also called adrenomedullin 2 (ADM2), is a newly discovered peptide related to calcitonin gene-related peptide (CGRP) and ADM. Many studies have shown that concentrations of ADM in cardiovascular tissue, plasma and urine were elevated in patients with acute coronary syndrome (ACS) [3,10,15]. Continuous administration of ADM had beneficial effects on remodeling of left ventricle and hemodynamics in rates with myocardial infarction, suggesting the possibility that ADM could be a useful therapeutic tool for acute myocardial infarction (AMI) [18] through counteracting renin–angiotensin–aldosterone system (RAAS) and oxidative stress [12,22]. It may also be an independent predictor of prognosis in patients with AMI [19]. IMD has significant vasodilator and cardiac protective actions similar to the related peptide ADM [21,24]. It exists extensively in the heart, kidney, hypothalamus, and plasma, and has a greater range of biological effects on the cardiovascular, respiratory and central nervous systems [17,24]. It dilates coronary arteries and enhances myocardial contraction [26] and reduces reactive oxygen species formation in vivo. The pathophysiological role of upregulated gene expression of AM2/IMD in the hearts of congestive heart failure (CHF) rats and increased plasma IMD levels in ischemia/reperfusion injury in isolated rat hearts seems to be related to its cardioprotective effects [8,27]. Recently, we discovered that exogenous administration of IMD could prevent the progression of atherosclerotic plaque [29]. Qin et al. [20] also found that the levels of IMD increased in patients with ACS. However, the dynamic tendency of IMD in patients with AMI is not clear, and whether it is a sensitive marker for the early diagnosis of coronary heart disease (CHD) remains unknown. The present study was designed to investigate the time course of plasma IMD levels and the potential role of IMD in AMI patients.

2. Materials and methods

2.1. Patients and study protocol

Twenty consecutive patients (75% men and 25% female; mean age 61.75 ± 13.521) were initially examined by ST-segment elevation myocardial infarction (STEMI) from the Intensive Care Unit (ICU) of West China Hospital Sichuan University within six hours following the onset of chest pain. We included a group of

* Corresponding author. Tel.: +86 028 85423352; fax: +86 028 85422175.
E-mail address: xiaopingc12@yahoo.cn (X. Chen).
[1] These authors contributed equally to this work.

thirty-three outpatients (77.4% men and 22.6% female; mean age 61.20 ± 5.029) with stable coronary heart disease (SHCD) from our hospital, and a group of eighteen control subjects from a community (age- and sex-matched), who had no previous diagnosis of hypertension, diabetes mellitus, other chronic diseases, and had not received any pharmacological therapy during the month prior to the study. AMI diagnosis was made using the following criteria [15]: (1) typical chest pain with a duration >30 min; (2) electro-cardiographic ST-segment elevation ≥0.1 mV in either two or more limb leads, or ≥0.2 mV in either two or more precordial leads; (3) and an elevation of serum creatine phosphokinase more than two times the upper limit of the normal range. All subjects provided written informed consent to participate in the study. The current protocol was approved by the Human Ethics Committee of the West China Hospital Sichuan University. Patients with renal failure (eGFR (estimated glomerular filtration rate) < 30 ml/min/1.73 m², malignancy, valvular heart disease, inflammatory diseases, and blood coagulation system disorders were excluded from the present study.

Peripheral venous blood samples for the measurements of IMD, malonaldehyde (MDA), superoxide dismutase (SOD) and cardiac biomarkers (myoglobin (Mb), creatine kinase MB (CK-MB) and cardiac troponin T (CTn-T)) were obtained from each patient with AMI at 12–24 h (day 1), 48 h (day 2), 96 h (day 4) and 168 h (day 7) following AMI, respectively. Blood samples from both SCHD and control subjects were collected for further assay. Plasma was separated using centrifugation at $3000 \times g$ for 15 min at 4 °C and was stored at −80 °C until extracted.

2.2. Peptide extraction and radioimmunoassay of plasma levels of IMD

Plasma IMD levels were assayed according to the method of Ryo Morimoto et al. [17]. Briefly, plasma samples were extracted with Sep-Pak C18 cartridges (Waters, Milford, MA, USA) and assayed by use of a radioimmunoassay kit (Phoenix Pharmaceuticals Inc., Belmont, CA, USA). Human $AM2/IMD_{1–47}$ (Peptide Institute) was used as a standard. $[^{125}I]$ $AM2/IMD_{1–47}$ (Phoenix) was used as a radioligand. The assay could detect changes of 3.3 ± 1.2 fmol/tube (mean ± SD; $n = 5$) from zero at 95% confidence with duplicate tubes. The crossreactivities were 100% with human IMD and less than 0.001% with other peptides including human ADM and CGRP. Intra- and interassay coefficients of variation were less than 5% and 10%, respectively.

2.3. Measurement of other variables

Fasting plasma glucose, serum lipids, brain natriuretic peptide (BNP) and myocardial markers were determined using standard laboratory measurements in our hospital laboratories. Lipid peroxidation was estimated by measuring TBARS according to a modified method of Jentzsch et al. [9]. Superoxide dismutase levels were measured using an ELISA assay, as previously described [1].

2.4. Documentation of coronary artery disease severity

During hospitalization, all patients with AMI underwent coronary angiography. The angiograms were assessed by two cardiologists who were unaware that the patients were to be included in the study. The coronary angiography score [25] was used to estimate the degree of coronary atherosclerosis. It was defined as the number of coronary branches (right, left anterior descending, and left circumflex) with more than 75% stenosis, with scores ranging from 0 to 3. We also excluded patients who only showed significance (more than 50% luminal obstruction) left of the main disease.

Table 1
Baseline characteristics of the study patients.

Risk factors	Groups		
	Control ($n = 18$)	SCHD ($n = 33$)	AMI ($n = 20$)
Age (years)	60.70 ± 2.21	61.20 ± 5.03	61.75 ± 13.52
Sex (male%)	60%	77.4%	75.0%
Hypertension	0%	70.9%	35.0%
DM history	0%	22.6%	20.0%
Smoking	10%	41.9%	40.0%
SBP (mmHg)	118.20 ± 10.40	122.80 ± 13.80	124.00 ± 17.49
DBP (mmHg)	69.50 ± 10.80	73.60 ± 7.09	73.30 ± 12.25
HR (bpm)	74.00 ± 6.00	74.10 ± 11.51	73.90 ± 12.99
BMI (kg/cm²)	22.84 ± 1.87	23.03 ± 2.66	24.01 ± 2.90
Cr (μmol/L)	73.44 ± 16.23	81.96 ± 22.08	88.51 ± 16.93
UA (μmol/L)	300.67 ± 39.75	369.91 ± 138.89	372.67 ± 88.46
FBG (mmol/L)	4.41 ± 0.56	5.72 ± 1.20*	7.40 ± 1.99*
TG (mmol/L)	1.64 ± 0.42	2.07 ± 0.63	1.91 ± 0.96
TC (mmol/L)	4.04 ± 0.94	3.63 ± 0.80*	4.07 ± 1.04*
HDL (mmol/L)	1.65 ± 0.29	1.21 ± 0.36*	1.26 ± 0.33*
LDL (mmol/L)	2.92 ± 0.78	2.46 ± 0.55*	3.23 ± 0.82*

* $P < 0.05$ versus control subjects.

2.5. Statistical analyses

Data are expressed as mean ± standard error of the mean, and all other variables are shown as a percentage of the control patients. Statistical significance of differences among groups was evaluated using ANOVA. The time course of plasma IMD, MDA, SOD and cardiac biomarkers was evaluated by analysis of variance for repeated measures. When the F value was found to be significant, the data were compared using a Dunnett's multiple comparison tests. Differences between the two groups were analyzed using either a χ^2 test or an unpaired Student's t-test. Differences were considered significant if $P < 0.05$.

3. Results

3.1. Patient characteristics

Baseline characteristics, including age, sex, body mass index (BMI), blood pressure, blood biochemical data, history of hypertension and diabetes mellitus among the patients with AMI, SCHD and the control subjects are listed in Table 1. We observed a large number of men with AMI (male:female = 3:1). The three groups did not differ significantly among age, gender, blood pressure, creatinine and uric acid, but the ANOVA showed significant differences in FBG, TC, HDL and LDL among the three groups. Statistical differences were also determined using pairwise comparison. TC and LDL were significantly lower in patients with SCHD than in controls ($P < 0.05$) and the AMI group ($P < 0.05$); patients with AMI had the highest FBG among the three groups ($P < 0.05$), and the SCHD patients had the lowest HLD ($P < 0.05$).

3.2. Levels of IMD, SOD, MDA, BNP and cardiac biomarkers in SCHD and control groups and the time course of these parameters in AMI patients

Plasma IMD, MDA and BNP levels in SCHD patients were significantly higher than in controls ($P < 0.05$), with a decrease in SOD activity in AMI patients ($P < 0.001$) when compared with the controls (Table 2). The time course of IMD, MDA, SOD and cardiac biomarkers is shown in Table 2, which reveals the variations in these indicators from the time of admission to the hospital to the seventh day following MI. At the time of admission to the hospital, patients with AMI exhibited elevated CTn-T levels compared to SCHD subjects ($P < 0.001$). The dynamic tendencies of CTn-T and