Clinical Studies

Myocardial and Peripheral Concentrations of β-Endorphin Before and Following Myocardial Ischemia and Reperfusion During Coronary Angioplasty

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SUMMARY

There is substantial evidence indicating that endogenous opioid peptides are involved in the pathophysiology of myocardial ischemia and reperfusion. We measured the myocardial and peripheral concentrations of β-endorphin before and following myocardial ischemia and reperfusion during coronary angioplasty. The results indicate that in patients with coronary artery disease, there was an augmented myocardial concentration of β-endorphin. Moreover, there was an increased peripheral concentration of β-endorphin following myocardial ischemia and reperfusion. The data support the previous notion that endogenous opioid peptides are involved in the pathophysiology of ischemic heart disease. (Jpn Heart J 2004; 45: 365-371)

Key words: Endogenous opioid peptides, Myocardial ischemia and reperfusion

THE endogenous opioid system includes three major families of peptides: dynorphin (derived from pre-proenkephalin B), endorphins (derived from pre-proopiomelanocortin), and enkephalins (derived from pre-proenkephalin A). Multiple forms of endogenous opioid peptides (EOP) are derived from these major precursors and many of them possess potent cardiovascular properties.

Since the identification of EOP and opioid receptors in the heart,[1] it has become clear that EOP subserve important roles in cardiovascular regulation and are especially involved in various cardiovascular stress situations such as shock,[2] heart failure,[3] and myocardial ischemia.[4]

In humans, it has been shown that opioid receptors are present in the heart and that they play an important role in myocardial ischemia.[5] We have shown that in patients with acute myocardial infarction, there was an increased plasma concentration of β-endorphin.[6] Mannheimer reported that there was a local myo-

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Received for publication May 6, 2003.
Revised and accepted November 13, 2003.

cardial release in the human heart of β-endorphin, leu-enkephalin, met-enkephalin, and dynorphin in patients undergoing pacing-induced angina pectoris.[7] On the other hand, it has been shown that myocardial infarction patients were associated with low γ-endorphin[8] or no change in β-endorphin levels.[9] Leu-enkephalin was found to be decreased in the myocardium of rats subjected to myocardial infarction.[10]

It is therefore of interest to investigate further whether or not there is direct release of β-endorphin in the human heart as well as concurrent increased plasma β-endorphin levels following myocardial ischemia and reperfusion.

METHODS

Consecutive patients scheduled for percutaneous transluminal coronary angioplasty (PTCA) with angiographically proven, functionally significant narrowing in one or more major coronary arteries were considered for entry into the study. After placement of the guiding catheter and performance of baseline coronary angiography, the view with the best visualization of the lesion was selected, and the severity of the coronary artery stenosis was measured using the stenosis diameter program included in the Philips DCI S System. Significant stenosis was defined as a reduction of > 70% of the lumen diameter. Informed consent was obtained before the PTCA procedure. Patients were excluded if they had one of the following: uncontrolled asthma or hypertension (blood pressure > 180/105 mmHg), acute myocardial infarction within 5 days, a left main artery stenosis of > 50%, or prior PTCA at the same site.

Clinical information, angiographic assessments, and intra- and postangioplasty complication data were recorded prospectively during hospitalization. PTCA was performed in the routine fashion. The choice of balloon type, inflation duration, and pressure were left to the operators. A 5F catheter was positioned in the coronary sinus before PTCA. The balloon was positioned across the stenosis and inflated as many times as needed to produce an optimal hemodynamic and angiographic result. Angiographic success was defined as a reduction in stenosis to < 30% residual narrowing after balloon dilatation associated with TIMI grade 3 flow. Blood samples were taken simultaneously from the coronary sinus and femoral artery through the coronary sinus catheter and a femoral arterial sheath, respectively. The samples were drawn before and after PTCA, as well as 6, 12, and 18 hours later. Determinations of serum β-endorphin, creatine kinase, CKMB, troponin, and lactate were performed. Blood samples were drawn into polyethylene syringes, placed immediately into iced test tubes containing EDTA as an anticoagulant, and centrifuged at 0°C for 15 minutes using 760 X g. Plasma obtained from the centrifuge was immediately frozen and stored at -70°C until

extraction and assay. For the extraction and determination of β-endorphin, a commercial radioimmunoassay kit was used.

All patients were taken to the intensive care unit after PTCA. If uneventful, the patients were transferred to an ordinary ward and subsequently discharged.

Data are expressed as the mean ± standard error of mean. Student's *t* test was used to test the differences in hemodynamic values, as well as in serum concentrations of creatine kinase and β-endorphin in the coronary sinus and femoral artery before and after the angioplasty, respectively. Analysis of variance (one-way ANOVA) was used to test the differences in serum concentrations of CKMB, troponin, and lactate before and after intervention. A P value less than 0.05 was considered statistically significant.

Results

A total of 9 consecutive patients with coronary artery disease (7 men and 2 women ranging in age from 58 to 83 years with a mean age of 70 ± 3 years) were studied. There were 4, 3, and 2 patients with one-vessel, two-vessel, and 3-vessel disease, respectively. Coronary angioplasty was performed successfully in all patients. All procedures were free of intra- and postangioplasty complications. All patients were subsequently discharged in good condition.

Table I illustrates the hemodynamic values obtained before and after the intervention. There were no significant differences in heart rate, arterial blood pressure, right atrial and pulmonary capillary wedge pressures, stroke volume, or cardiac output before and after angioplasty.

Figure 1 shows the plasma levels of CKMB, troponin, and lactate before and 6, 12 and 18 hours after the PTCA procedure. Creatine kinase levels were 92 ± 24 and 209 ± 76 U/L before and after the intervention, respectively ($P = 0.21$). There were no significant differences in the plasma levels of creatine kinase, CKMB, troponin, and lactate before and following angioplasty.

Table II shows the concentrations of β-endorphin in the coronary sinus and femoral artery before and after the PTCA procedure, respectively. Before angio-

Table I. Hemodynamics Before and After Coronary Angioplasty

	Before angioplasty	After angioplasty
Heart rate (beats/min)	79.11 ± 4.99	76.00 ± 3.79
Mean arterial pressure (mmHg)	106.30 ± 5.86	102.41 ± 4.94
Right atrial pressure (mmHg)	9.13 ± 0.85	10.63 ± 1.71
Pulmonary capillary wedge pressure (mmHg)	19.88 ± 2.15	20.50 ± 2.13
Cardiac output (L/min)	4.69 ± 0.35	3.98 ± 0.31
Stroke volume (mL/beat)	59.59 ± 5.03	53.86 ± 5.50

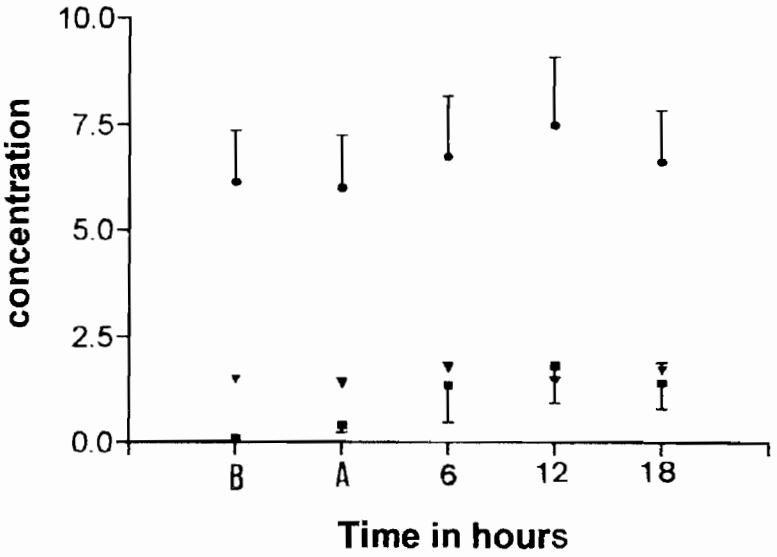

Figure 1. Plasma concentrations of CKMB in U/L (●), troponin in μg/ml. (■) and lactate in mmole/l. (▼) before (B), immediately after (A), and 6, 12, and 18 hours following coronary angioplasty.

Table II. β-endorphin Concentrations in Coronary Sinus and Femoral Artery Before and After PTCA

	Before PTCA	After PTCA
Coronary sinus	75.21 ± 16.72 ★	32.72 ± 6.44
Femoral artery	15.19 ± 3.75	48.77 ± 9.74*

Values are means (SEM) in pmole/L. ★ $P < 0.05$ coronary sinus vs femoral artery; *$P < 0.05$ before vs after PTCA.

plasty, the mean β-endorphin levels were 75.21 ± 16.72 and 15.19 ± 3.75 pmole/L in the coronary sinus and femoral artery, respectively. Following the intervention, however, the β-endorphin concentrations became 32.72 ± 6.64 pmole/L in the coronary sinus and 48.77 ± 9.74 pmole/L in the femoral artery. There were no changes in β-endorphin concentrations in the coronary sinus ($P = 0.35$) but there was a significant increase in β-endorphin level in the femoral artery ($P < 0.05$) following the PTCA procedure. Moreover, there was a significantly higher concentration of β-endorphin in the coronary sinus than in the femoral artery before the intervention ($P < 0.05$).

DISCUSSION

EOP and opioid receptors are widely distributed throughout the body.[11] Since the identification of EOP and opioid receptors in the heart,[1] it has become clear that EOP subserve important roles in various cardiovascular stress situations such as shock and myocardial ischemia, contributing to the respective deleterious effects.[2-4] There is substantial evidence that cardiac opioid receptors are activated during myocardial ischemia and reperfusion, supporting the hypothesis that EOP are involved in the pathophysiology of ischemic heart disease.[12] We have shown that in patients with acute myocardial infarction, there was an increased plasma concentration of β-endorphin.[6] In this study, we have further demonstrated that in patients with coronary artery disease, there was an elevated myocardial concentration of β-endorphin, since there was a higher concentration of β-endorphin in the coronary sinus than in the femoral artery. The results are compatible with those of Mannheimer[7] and Bagrov,[13] but not with those of Dmitrieva,[9] Bernardi,[10] and Maslov.[11] The discrepancy may be due to differences in experimental protocols. However, whether or not there is any difference in β-endorphin levels between normal cases and coronary artery disease cases warrants further studies.

In this study, there were no significant differences in hemodynamic values or plasma levels of creatine kinase, CKMB, troponin, and lactate before and after the PTCA procedure. This indicates that the coronary angioplasty was performed smoothly and successfully without causing any myocardial damage in our patients.

The widespread distribution and localization of opioids and receptors throughout the cardiovascular system suggest that the opioid system can be activated by the cardiovascular stress situation due to myocardial ischemia and reperfusion locally or systematically.[11] In this study, there were no changes in β-endorphin concentrations in the coronary sinus before and after PTCA. But there was a significant increase in the β-endorphin level in the femoral artery after PTCA. The results suggest that the cardiovascular stress situation due to myocardial ischemia and reperfusion following PTCA leads to an increased systemic release of β-endorphin in the general circulation, but not a local myocardial release in the heart. Thus, the myocardial and plasma contents of β-endorphin may be differently controlled and originate from different sources. However, since there is increased basal myocardial content of β-endorphin in patients with ischemic heart disease, we must also consider the possibility that further myocardial ischemia and reperfusion (by PTCA) may not further increase the local myocardial release of β-endorphin in the adapted heart. Moreover, there may be an exhaustion of the opioid system at the level of the myocardium in coronary artery

disease patients with an augmented endogenous opioid tone. Or the ischemic stress situation may be alleviated locally following revascularization of the coronary lesions. Further studies are needed to clarify this. Nevertheless, the results are compatible with the hypothesis that, during myocardial ischemia and reperfusion, there is activation of the endogenous opioid system causing release of EOP, suggesting that EOP play an important role in ischemic heart disease.

There are several pieces of evidence suggesting that EOP are involved in myocardial ischemia. Firstly, isolated perfused hearts of rats chronically treated with morphine exhibit tolerance to dynorphin or ischemia and reperfusion in induction of arrhythmias.[14] Secondly, the opioid antagonists naloxone,[15] naltrexone,[16] and MR2266[17] have been shown to attenuate arrhythmias during ischemia and reperfusion in the isolated rat heart. Thirdly, the antiarrhythmic effects of naloxone are stereospecific because the (+) isomer of naloxone (without any opiate antagonistic property) is not effective in reversing ischemia-induced arrhythmias, hypotension, or bradycardia in the rat.[18] Fourthly, dynorphin and naloxone potentiated and attenuated, respectively, ischemia induced arrhythmias, bradycardia, and cardiogenic shock in rats subjected to coronary artery ligation.[19] The above strongly indicate that EOP are involved in the pathophysiology of myocardial ischemia. Though there are limitations in this study population with a limited number of patients (this was due to the trial design), the striking results of this initial study, however, clearly indicate that in patients with coronary artery disease, there were elevated levels of myocardial β-endorphin and increased peripheral concentrations of β-endorphin following myocardial ischemia and reperfusion, supporting the previous notion that EOP may be involved in the pathophysiology of ischemic heart disease. Further investigation is needed to identify the types of EOP that are released, and to elucidate the events that occur following myocardial ischemia and reperfusion.

REFERENCES

1. Krumins SA, Faden AL, Feuerstein G. Opiate binding in rat hearts: modulation of binding after hemorrhagic shock. Biophys Res Commun 1985; 127: 120-8.
2. Holaday JW. Cardiovascular consequences of endogenous opiate antagonism. Biochem Pharmacol 1983; 32: 573-83.
3. Liang CS, Imai N, Stone CK, Woolf PD, Kawashima S, Tuttle RR. The role of endogenous opioids in congestive heart failure: effects of nalmefene on systemic and regional hemodynamics in dogs. Circulation 1987; 75: 443-51.
4. Fagbemi O, Lepran I, Parratt JR, Szekeres L. Naloxone inhibits early arrhythmias resulting from acute coronary ligation. Br J Pharmacol 1982; 76: 504-6.
5. Tomai F, Crea F, Gaspardone A, *et al*. Effects of naloxone on myocardial ischemic preconditioning in humans. JACC 1999; 33: 1863-9.

6. Chen YT, Lin CJ, Lee AYS. Plasma levels of endogenous opioid peptides in patients with acute myocardial infarction. Jpn Heart J 1995; 36: 421-7.
7. Mannheimer C, Emanuelsson H, Larsson G, *et al*. Myocardial release of endogenous opioids in the human heart and the effects of epidural spinal electrical stimulation in pacing-induced angina pectoris. JACC 1991; 17: 107A.
8. Dmitrieva OF, Ivanikow IO. Blood endorphin levels in myocardial infarction. Kardiologiia 1987; 27: 62-5.
9. Bernardi P, Fontana F, Pich EM, Spampinato S, Canossa M. Plasma endogenous opioid levels in acute myocardial infarction patients with and without pain. Eur Heart J 1992; 13: 1074-9.
10. Maslov LN, Lishmanov YB. Change in opioid peptide level in the heart and blood plasma during acute myocardial ischemia complicated by ventricular fibrillation. Clin Exp Pharmacol Physiol 1995; 22: 812-6.
11. Hughes J, Kosterlitz HW, Smith TW. The distribution of methionine-enkepahlin and leucine-enkephalin in the brain and peripheral tissues. Br J Pharmacol 1977; 61: 639-47.
12. Lee AYS. Endogenous opioid peptides and cardiac arrhythmias. Int J Cardiol 1990; 27: 145-51.
13. Bagrov LY. Plasma β-endorphin levels in acute myocardial infarction. Ann Emerg Med 1993; 22: 152-3.
14. Wong TM, Lee AYS. Chronic morphine treatment reduces the incidence of ventricular arrhythmias in the isolated rat heart induced by dynorphin or myocardial ischemia and reperfusion. Neurosci Lett 1987; 77: 61-5.
15. Zhan CY, Lee AYS, Wong TM. Naloxone blocks the cardiac effects of myocardial ischemia and reperfusion in the isolated rat heart. Clin Exp Pharmacol Physiol 1985; 52: 373-8.
16. Liu XL, Mok CP, Lee AYS, Wong TM. Evaluation of antiarrhythmic potency of naltrexone in isolated ischemic rat heart. Acta Pharmacol Sin 1988; 9: 40-3.
17. Wong TM, Lee AYS, Tai KK. Effects of drugs interacting with opioid receptors during normal perfusion or ischemia and reperfusion in the isolated rat heart — An attempt to identify cardiac opioid receptor subtypes involved in arrhythmogenesis. J Mol Cell Cardiol 1990; 22: 1167-75.
18. Kan MN, Chen YT, Lee AYS. Naloxone reversal of ischemic arrhythmias is stereospecific and suggests role of endogenous opioid peptides in ischemic heart disease. Proc Soc Exp Biol Med 1992; 200: 518-21.
19. Wu JP, Chen YT, Lee AYS. Opioids in myocardial ischemia: potentiating effects of dynorphin on ischemic arrhythmias, bradycardia and cardiogenic shock following coronary artery occlusion in the rat. Eur Heart J 1993; 14: 1273-7.