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The Blocking Effects of Nalmefene --- A new, Potent, and Long-Lasting Opiate Antagonist --- Against Myocardial Ischemia-Induced Arrhythmias in Rats

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Background. It has been shown that cardiac opioid receptors are activated during arrhythmias induced by myocardial ischemia, supporting the hypothesis that endogenous opioid peptides (EOPs) are involved in cardiac arrhythmogenesis and in myocardial ischemia. The opiate antagonist naloxone has been found to inhibit cardiac arrhythmias resulting from coronary artery occlusion in both in vitro and in vivo preparations. It is therefore of interest to further investigate whether another potent, long-lasting opiate antagonist, nalmefene, also blocks the cardiac effects of myocardial ischemia.

Methods and Results. Spraque-Dawley rats were anesthetized and artificially ventilated. Left thoracotomy was performed and the left coronary artery was ligated. Following coronary artery occlusion, all rats in the control group developed ischemia-induced arrhythmias, bradycardia and hypotension, which were significantly inhibited by high dose pre-treatment with nalmefene.

Conclusions. The results indicate that EOPs may be released when myocardial ischemia occurs, thus causing arrhythmias, bradycardia and hypotension. Nalmefene, by virtue of its antagonistic action, attenuates these fatal complications secondary to myocardial ischemia. This suggests that EOPs play an important role in ischemic heart disease and that nalmefene may have therapeutic value.

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Key Words: Opiate antagonist, Nalmefene, Myocardial ischemia, Endogenous opioid peptides

Introduction

It has been shown that endogenous opioid peptides (EOPs) are involved in cardiac arrhythmogenesis [1] and that the opiate antagonists naturally possess antiarrhythmic activity[2]. The opiate antagonist naloxone has been found to inhibit cardiac arrhythmias resulting from coronary artery occlusion in rats[3,4] and dogs[5], suggesting that EOPs may be released from the heart upon myocardial ischemia thus causing arrhythmia, and that naloxone, by virtue of its antagonistic action against opiates, rectifies this irregular cardiac rhythm.

It has been suggested therefore that opiate antagonists may be used as antiarrhythmic agents. Naloxone, being the first pure opiate antagonist and most commonly used, was naturally the first one to be considered. Its antiarrhythmic potency was determined with a screening test using the isolated ischemic perfused rat heart preparation developed by us.[4] It was found to be comparable to those of the prototype antiarrhythmic agents, namely, lidocaine,

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quinidine and propranolol.[6] However, naloxone is short-acting, with a half-life in humans of about one hour, [7]and is easily degraded when administered orally. Nalmefene, (5α)-17-(cyclopropylmethyl)-4,5-epoxy-6-methylenemorphinan-3,14-diol, another pure opiate antagonist with a much longer half-life of 8-9 hours in man, is reported to be 18 times more potent than naloxone.[8] Nalmefene seems capable of potent, long-lasting opiate antagonistic effects and thus was examined for its blocking effects against myocardial ischemia-induced arrhythmias in the present study.

Methods

Sprague-Dawley rats of either sex weighing between 350 to 400 gm were used. All experiments were conducted according to guidelines for animal experiments at Taichung Veterans General Hospital Medical Research Center. Rats were anesthetized with sodium pentobarbitone (60 mg.Kg^{-1}) intraperitoneally. A tracheotomy was performed, then the intubated cannula was connected to a rodent ventilator (Harvard Apparatus, Massachusetts, USA) and the rats were ventilated artificially with room air (60-80 strokes min^{-1} 1 ml. 100 gm^{-1}). The left femoral artery and vein were cannulated for the measurement of blood pressure and heart rate by a Statham pressure transducer and a Biotechnometer (Gould) and for the administration of drugs, respectively. Electrocardiograms were recorded from lead II limb leads, with a positive electrode connected to the left leg, a negative electrode to the right arm, and a ground electrode to the left arm. The Lifepak ECG Monitor (Physio-Control Corp, USA) was used for all electrocardiographic recordings. When cardiac arrhythmias such as ventricular premature contraction (VPC) and ventricular tachycardia (VT) occurred, heart rate was measured from the electrocardiogram by averaging the RR intervals. When ventricular fibrillation (VF) occurred and the heart was fibrillating but not beating, heart rate became zero as recorded by the Biotechnometer.

Left thoracotomy in the fifth intercostal space was performed. The heart was exposed and a ligature (6/O silk suture) was placed around the left coronary artery. The rat was then allowed to equilibrate for 15 minutes. During this period, any rat showing functional instability such as hypotension or occurrence of cardiac arrhythmias was discarded. After stabilization, nalmefene (8 umole Kg^{-1} and 24 umole Kg^{-1}) or saline as control were injected intravenously 10 minutes before the ligature was tied. Blood pressure, heart rate and electrocardiogram were then continuously monitored throughout the thirty minute' post-ligation period. To enable quantitative comparison, an arrhythmia scoring system adopted from Curtis and Walker[9] was used. In this study, both arrhythmia scores and raw arrhythmia data were presented. Each rat was given one score, representing the most severe type of arrhythmias observed during the entire post-ligation period. Details of the scoring system were: score 0 = no arrhythmia; score 1 = occasional VPC; score 2 = frequent VPC; score 3 = VT, 1-2 episodes; score 4 = VT, 3-5 episodes; score 5 = VT, > 5 episodes; score 6 = VF, 1-2 episodes; score 7 = VF, 3-5 episodes; score 8 = VF, > 5 episodes.

A chi-squared test was used to analyze the difference in the incidence of arrhythmias between control and treated groups. Student's t test was used to test the difference in arrhythmia scores and in the onset of arrhythmias between control and treated groups. Analysis of variance was used to compare the difference in time course changes in mean arterial pressure and heart rate between control and treated groups. A p value of less than 0.05 was considered as statistically significant.

Results

Table 1 summarizes the effects of nalmefene on the cardiac rhythm following coronary artery occlusion. Myocardial ischemia invariably caused ventricular arrhythmias, including VPC, VT and VF. Following coronary artery ligation, all rats in the control group developed ischemic arrhythmia in the 30 minutes post-ligation period. Of eight rats, eight showed VPC, seven had VT and four developed VF. The onset of arrhythmias were at 3.80, 5.70 and 7.75 minutes, respectively. The overall arhythmia score was 5.38. Of eight rats receiving 8 umoleKg^{-1} nalmefene, eight showed VPC, six had VT and five developed VF. The onset of arrhythmias were at 6.25, 8.67 and 10.2 minutes, respectively. The mean arrhythmia score was 5.63. At a higher dose, 24 umoleKg^{-1} of nalmefene, six out of eight rats showed VPC, three had VT and none developed VF. The onset of arrhythmias were at 5.00 and 5.00 minutes, respectively. The overall arrhythmia score was 2.00, which was significantly lower than that of the control group. Thus, pretreatment with nalmefene at the dose of 24 umoleKg^{-1} before coronary artery ligation significantly attenuated the incidence and severity of the

Table 1. Effects of nalmefene on cardiac rhythm following coronary artery occlusion in rats.

	N	Arrhythmia score	n	VPC onset(min)	n	VT onset(min)	n	VF onset(min)
Control	8	5.38±0.80	8	3.50±0.91	7	5.71±0.84	4	7.75±1.03
Nalmefene (8 umoleKg^{-1})	8	5.63±0.81	8	6.25±1.35	6	8.67±1.76	5	10.20±0.78*
Nalmefene (24umoleKg^{-1})	8	2.00±0.60**	6	5.00±2.04	3⋆	5.00±2.89	0	

n = number of rats. VPC = ventricular premature contraction; VT=ventricular tachycardia; VF= ventricular fibrillation. *,**P < 0.05 and < 0.01 vs corresponding control group (student's t test); ⋆P < 0.05 vs control group (chi-square test).

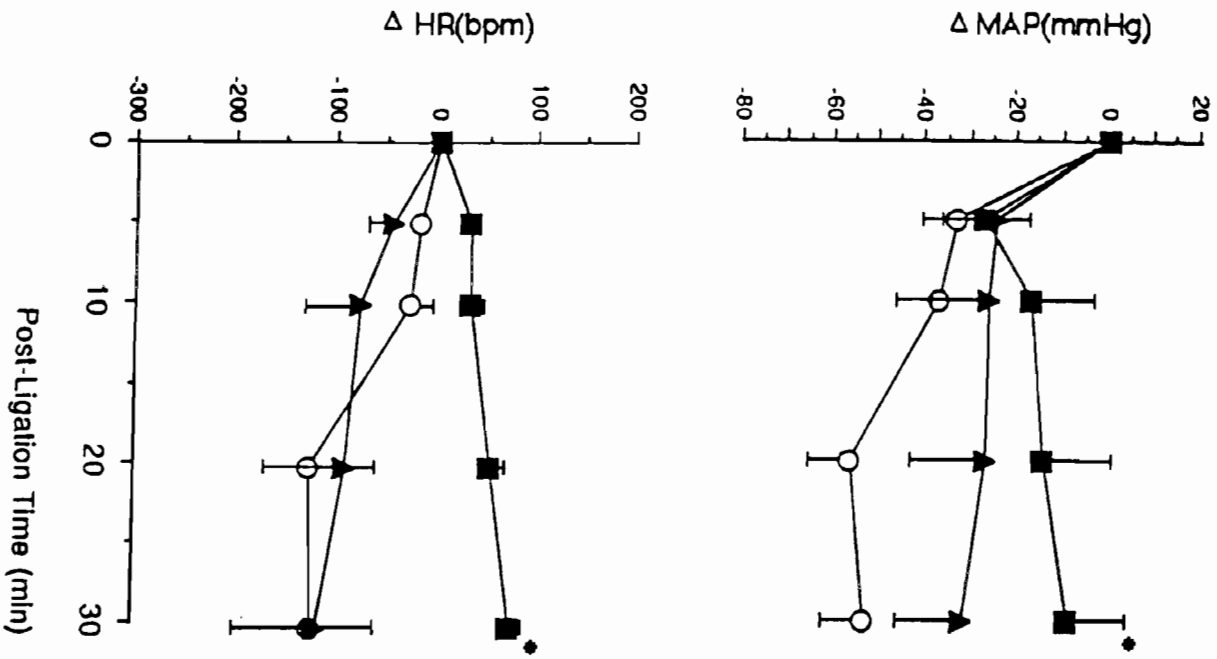

Figure 1. Effects of nalmefene on change in mean arterial pressure (△MAP, upper tracing) and mean heart rate (△HR, lower tracing) following coronary artery occlusion in rats. (○) saline; (▲) nalmefene 8 umole.Kg^{-1}; (■) nalmefene 24 umole.Kg^{-1}. Values are means, bars = SEM, of eight animals. *P < 0.05 vs control by analysis of variance.

ischemia-induced arrhythmias.

Figure 1 shows the effects of nalmefene on the changes in mean arterial blood pressure and heart rate following coronary artery occlusion. Myocardial ischemia invariably caused a marked decrease in both arterial pressure and heart rate. Following coronary artery ligation, there were profound reductions in both mean arterial pressure and heart rate in the control group. Pretreatment of nalmefene at the dose of 24 umoleKg^{-1} before coronary artery ligation, however, significantly attenuated ischemia-induced hypotension and bradycardia.

Discussion

It is well known that myocardial ischemia leads to cardiogenic shock, bradycardia, and ischemic arrhythmias. Similar observations were made in the present study in which coronary artery occlusion soon led to malignant ventricular arrhythmias, and a marked reduction in arterial blood pressure and heart rate. In this study, nalmefene, a potent and long-lasting opiate antagonist, reduced the incidence and severity of ischemia-induced arrhythmias, bradycardia and cardiogenic shock, indicating that nalmefene blocked the cardiac effects of myocardial ischemia in the rat. The results are in agreement with those reported previously on the antiarrhythmic effect of naloxone,[3,5,10] and also support the notion that EOPs may be involved in arrhythmogenesis of the heart. According to Sawynork et al,[11] more compelling proof of a role of EOPs in any situation requires several lines of evidence that include the production of similar responses with various opiate antagonists. Thus, the antiarrhythmic action of nalmefene as well as of other opiate antagonists such as naloxone,[3,5,10] naltrexone,[12] and MR2266[13] can be explained on the basis that blockade of opioid receptors inhibits ischemia induced arrhythmias by reducing the effects of EOPs released as a consequence of the stress of myocardial ischemia.

EOPs and their receptors have been found in the heart.[1] This opioid system plays important roles in cardiovascular regulation and is involved in the pathophysiology of myocardial ischemia. Administration of opioid agonists has been demonstrated to potentiate ischemia-induced arrhythmias, bradycardia and hypotension, and these effects are prevented by the opiate antagonists.[14,15] Results of the present study also show that nalmefene limited and delayed the occurrence of ischemic events secondary to coronary arterial ligation, which further infer that EOPs are factors in myocardial ischemia. The above are also consistent with the hypothesis that myocardial ischemia activates the myocardial opiate receptors through an increased release of EOPs from the heart, thereby causing arrhythmias, bradycardia and hypotension. By virtue of their antagonistic actions against opiates, opiate antagonists can attenuate these ischemic events, thus suggesting that EOPs play an important role in ischemic heart disease.

The antiarrhythmic effects of opiate antagonism have considerable clinical implications in the prevention and treatment of cardiac arrhythmias and ischemic heart disease. Further studies are needed to define the extent of involvement of EOPs in cardiac arrhythmogenesis and to elucidate both the mechanisms of action of EOPs in the pathophysiology of myocardial ischemia and the electrophysiological effects of opioid receptor blockade in cardiac muscle. More studies are also warranted to define the therapeutic values of opiate antagonists.

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Nalmefene 在心肌缺血時之作用

李應紹

背景：已有報導，在心肌缺血時，心臟鴉片受體會被激活而產生心律不整。而鴉片對抗劑 Naloxone 則能抑制冠狀動脈結紮時產生之心律異常。本研究在進一步探討另一種強而長效的鴉片對抗劑 Nalmefene，在心肌缺血時的作用。

方法：大白鼠經麻醉後進行人工呼吸。左邊開胸後結紮左冠狀動脈；比照組在冠狀動脈結紮後均產生心律不整，低心跳及低血壓。而Nalmefene 則能抑制此等心肌缺血時產生之異常。

結論：研究結果顯示心肌缺血會導致內生性鴉片樣胜之釋出，從而引發心律不整，低心跳及低血壓。Nalmefene 具抑制此等心肌缺血時產生之併發症。Nalmefene 可能對缺血性心臟病及心律不整具療效。

關鍵詞：鴉片對抗劑，Nalmefene，心肌缺血，內出性鴉片樣胜

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