β-Endorphin, an endogenous depressor agent in the rat?

M. A. PETTY, J. M. A. SITSEN AND W. DE JONG
Rudolf Magnus Institute for Pharmacology, Medical Faculty, University of Utrecht, Utrecht, The Netherlands

Summary
1. The role of opiates in cardiovascular regulation has been investigated.
2. In urethane-anaesthetized renal hypertensive rats (two-kidney, one-clip Goldblatt model), intracerebroventricular β-endorphin (10 μg) caused a greater fall in mean arterial pressure than in sham-operated controls.
3. Unilateral injection of β-endorphin into the nucleus tractus solitarii of the urethane-anaesthetized rat resulted in a U-shaped dose-response relationship, with a fall in mean arterial pressure and heart rate occurring at low doses. Doses above 10 ng caused a rise in pressure, accompanied by a variable effect on heart rate.
4. The fall in blood pressure and heart rate was prevented by prior subcutaneous administration of naloxone. Naloxone caused an increase in blood pressure when administered alone.
5. These results suggest a depressor role of an endogenous brain opiate, possibly β-endorphin; a site of action is probably the nucleus tractus solitarii.

Key words: β-endorphin, nucleus tractus solitarii, opiates.

Abbreviations: HR, heart rate; MAP, mean arterial pressure; NTS, nucleus tractus solitarii.

Introduction
Anatomical and pharmacological evidence suggests a role for endogenous opiates in central cardiovascular regulation. Two different opiate systems exist, the enkephalins, which are distributed throughout the brain with numerous cell groups and short neuronal connections, and, secondly, the endorphin system. The latter is located in a single set of hypothalamic neurons, with axons projecting throughout the brain [1]. More specifically, enkephalinergic neurons and their terminals, together with binding sites for opiates, have been localized in brain-stem structures involved in cardiovascular control [2, 3].

The intracisternal administration of opioid peptides results in a decrease in baroreflex sensitivity [4] and variable effects on blood pressure and heart rate. Leu-enkephalin, Met-enkephalin and the stable analogue D-Ala-enkephalin cause a rise in blood pressure on central administration [5, 6]. Conversely, morphine and β-endorphin produce an initial rise in pressure followed by a prolonged hypotensive phase [6, 7]. Other authors observed only a depressor response [8]. All these effects are readily reversed by the opiate antagonist naloxone.

Intravenous and central naloxone causes a rise in blood pressure in different shock states, implicating the involvement of endogenous opiates in central cardiovascular control [9, 10, 11], and in particular a depressor role of endorphins.

Furthermore Farsang and co-workers reported that naloxone reverses the hypotensive effect of clonidine and α-methyldopa in spontaneously hypertensive rats [12]. In addition, clonidine has been reported to induce the release of a β-endorphin-like peptide from brain-stem slices [13]. These observations point to a depressor role of β-endorphin.

Recently, Zamir et al. reported an altered pain sensitivity in hypertensive rats and in essential hypertensive patients. Administration of naloxone to the hypertensive rats reversed the increased pain threshold (hot plate latency) to control values [14, 15, 16]. These data suggest that under these circumstances there is a change in endorphin systems.

In this study the cardiovascular effects of β-endorphin injected into the lateral cerebral
ventricle in renovascular hypertensive rats and sham-operated controls have been examined.

Freye & Arndt [17] have indicated that opiate binding sites bordering the fourth cerebral ventricle mediate the hypotension and bradycardia observed after fentanyl perfusion. The possibility that the nucleus tractus solitarii (NTS) is a site of action in mediating the cardiovascular effects of opiates and in particular β-endorphin has been investigated.

Methods

Male Wistar rats (Cpb, TNO, Zeist, The Netherlands) weighing 200–250 g were used.

Renal hypertension was induced by the application of a solid silver clip (0.20 mm internal diameter) to the left renal artery of rats weighing 125–135 g (two-kidney, one-clip Goldblatt model). The control group was subjected to the same operative procedure, but no clip was applied. The rats were used 3–4 weeks after operation.

β-Endorphin (10 μg) or an equivalent volume of artificial cerebrospinal fluid vehicle was injected into the right lateral brain ventricle of urethane-anaesthetized (1.5 g/kg) renovascular hypertensive rats and sham-operated controls by means of a cannula implanted 1 week earlier under Hypnorm anaesthesia (Duphar Nederland BV, Amsterdam, The Netherlands).

For direct application of β-endorphin into the NTS the rat was placed in a stereotactic apparatus and the head fixed to an angle of 45° downward. After exposure of the lower brain stem micro-injections into the NTS on the right side of the medulla were given through a glass cannula (outer diameter 60 μm), in a volume of 0.4 μl, which was delivered in 10 s [18]. After experimentation the correct position of the injection site was confirmed by histology.

The following drugs were used: β-endorphin (Organon International BV, Oss, The Netherlands) dissolved in artificial cerebrospinal fluid for intracerebroventricular injection and in 0.9% sodium chloride solution (saline) for direct application into the NTS; naloxone/HCl (Endo Laboratories, New York, U.S.A.) in saline vehicle.

Results

The renal hypertensive rats had a significantly \( (P < 0.001) \) higher mean arterial pressure (MAP) \( (162 \pm 8 \text{ mmHg}) \) than the sham-operated controls \( (100–120 \text{ mmHg}) \). Intracerebroventricular administration of β-endorphin (10 μg) in the hypertensive group resulted in a significantly \( (P < 0.05) \) greater fall in MAP than in controls. In both groups the fall was delayed, occurring 15 min after injection, when there was a gradual decrease in MAP, which reached a maximum at 60 min, 37 ± 34 mmHg and 9 ± 20 mmHg respectively. No significant changes in heart rate (HR) or respiratory frequency were observed.

The unilateral injection of β-endorphin into the NTS of normotensive animals resulted in a U-shaped dose–response relationship with a fall in MAP and HR occurring at low doses (1–1000 pg). The fall in both MAP and HR occurred gradually over the following 60 min, the changes

![Fig. 1](image-url)  

**Fig. 1.** Specimen traces (a) of the hypotension and bradycardia resulting from β-endorphin (100 pg) injected unilaterally into the nucleus tractus solitarii of the urethane-anaesthetized rat, (b) after injection of saline control.Injection volume was 0.4 μl.
Cardiovascular effects of β-endorphin

respectively (Fig. 1). No change in respiratory frequency was observed. Both the hypotension and bradycardia were prevented (P < 0.001) by the administration of naloxone (1 mg/kg, subcutaneously) given 15 min before the peptide. Naloxone caused an increase in MAP of 8 ± 3 mmHg, when administered subcutaneously, followed 10 min later by a saline injection into the NTS. The maximum effect occurred 40 min after naloxone administration.

Doses of β-endorphin greater than 10 ng caused a rise in MAP accompanied by variable effects on HR when administered into the NTS. The maximum rise in pressure was observed after 30 min. Baseline values were reached 40–60 min after injection. No decrease in MAP was seen.

Discussion

In this study the role of β-endorphin in central cardiovascular regulation has been investigated. The intracerebroventricular administration of the opiate peptide caused a greater fall in blood pressure in renal hypertensive rats than in sham-operated controls. This observation indicates the alteration of an endogenous opiate system in hypertension. Zamir et al. [14] have shown a 45% higher level of 'opioid activity' in the spinal cord of renal hypertensive animals than in controls.

It has been postulated that opiate receptors bordering the fourth cerebral ventricle mediate the hypotensive and bradycardiac effects of some opiates [17]. The nucleus tractus solitarii (NTS) is the site of the first synapse in the baroreflex arc and it has been shown that opiate peptides affect baroreceptor reflex function [4]. Therefore, does β-endorphin lower blood pressure through an action in the NTS? The local application of the peptide into this region resulted in a U-shaped dose–response relationship with hypotension accompanied by bradycardia occurring at low doses. (As seen in Fig. 1, the decrease in blood pressure occurs gradually during a period of 1 h.) At doses above 10 ng a rise in pressure was observed, perhaps due to the simultaneous activation of other opiate receptors with a lower affinity for β-endorphin or by diffusion to opiate receptors inaccessible to lower doses. The depressor effect appears to be due to the activation of opiate receptors since it was antagonized by naloxone. Naloxone treatment in the absence of β-endorphin caused a significant rise in blood pressure, as also shown by Kumazawa et al. [19]. Preliminary studies involving the local application of antiserum to β-

endoorphin into the NTS show a delayed but significant rise in pressure.

These findings suggest a depressor role of β-endorphin in the central nervous system, very low doses causing a fall in blood pressure when administered into the NTS. It is possible that the hypotension results from an action within the central connections of the baroreflex arc. These findings and data from the literature give evidence that β-endorphin may be involved in central blood pressure regulation, in both hypertension and shock states, as well as in normotension.

Acknowledgments

We thank Mr H. de Lang for his excellent technical assistance. M.A.P. is supported by a grant from the British and Dutch Heart Foundations.

References


