Effects of enkephalins on arterial blood pressure are reduced by propranolol

Departments of Pharmacology and Physiology, University of Heidelberg, Heidelberg, Germany

Summary

1. The cardiovascular effects of enkephalins have been tested in normotensive Wistar-Kyoto rats. Methionine-enkephalin and leucine-enkephalin increased blood pressure and heart rate after infusion into the brain ventricles.

2. After intravenous injection, blood pressure was increased by methionine-enkephalin and leucine-enkephalin, but heart rate was increased by methionine-enkephalin only.

3. Propranolol treatment reduced the increases in blood pressure following intraventricular methionine-enkephalin and leucine-enkephalin, while only the methionine-enkephalin-induced increases in heart rate were reduced by propranolol.

4. Heart rate and blood pressure responses after intravenous administration of methionine-enkephalins and leucine-enkephalin were not affected by propranolol.

5. Since opioid peptides occur in the blood and in regions of the brain involved in blood pressure regulation, the demonstrated cardiovascular effects to intraventricular and intravenous enkephalins support a role of these peptides in central and peripheral mechanisms of blood pressure control.

Key words: blood pressure, brain, heart rate, leucine-enkephalin, methionine-enkephalin, propranolol.

Abbreviations: Leu-ENK, leucine-enkephalin; Met-ENK, methionine-enkephalin.

Introduction

Recently, peptides have been discovered in various parts of the brain which were previously thought to occur in peripheral tissue or in one distinct region of the brain only (Hökfelt, Elde, Johansson, Ljungdahl, Schultzberg, Fuxe, Goldstein, Nilson, Pernow, Terenius, Ganten, Jeffcoat, Rehfeld & Said, 1978; Hughes, Smith, Kosterlitz, Fothergill, Morgan & Morris, 1975). Thus, angiotensin II which has long been known as a hormone circulating in blood, has been shown to occur also in the brain. Both circulating humoral angiotensin II and brain angiotensin II appear to subserve blood pressure regulation (Ganten, Fuxe, Phillips, Mann & Ganten, 1978). Morphinomimetic peptides also occur in blood and in brain tissue. Their localization in the nucleus tractus solitarius, amygdaloid nucleus, hypothalamus and median eminence suggest that enkephalins might be involved in central blood pressure control and neuroendocrine regulation.

It has previously been demonstrated that part of the central angiotensin II effect on blood pressure is due to release of anti-diuretic hormone from the hypophysis, and there is now evidence that anti-diuretic hormone secretion can also be stimulated by opioid peptides (Bisset, Chowdrey & Feldberg, 1978). Furthermore, opioid peptides and adrenocorticotropin hormone appear to originate from one common precursor and are secreted in equimolar quantities into the blood (Guillemin, Vargo, Rossier, Minick, Luig, Rivier, Volp & Bloom, 1977). Both are secreted in stressful situations such as pain (Guillemin et al., 1977). If enkephalins had an effect on blood pressure, they could contribute to the haemodynamic adaptation occurring in stressful situations (Barker & Smith, 1977). In the
following experiments we examined the blood pressure responses to leucine-enkephalin and methionine-enkephalin after infusion into the brain ventricles and after intravenous injection. We also tested the effect of propranolol on the blood pressure responses to enkephalins, since it has been shown that propranolol reduces central peptide effects (Johnson, Simon, Schaz, Ganten, Ganten & Mann, 1978).

Methods

Adult male normotensive Wistar-Kyoto (WKY) rats, body weight 300–350 g, were used in all experiments.

Testing procedures

A chronic 23 gauge guide cannula was stereotactically implanted into the lateral ventricle of the brain for infusion of the test substances. The method of brain cannulation has been described in detail elsewhere (Mann, Phillips, Dietz, Haebara & Ganten, 1978). After the operation, the rats were allowed a minimum of 6 days to recover. On the day of testing catheters were implanted under ether anesthesia into the femoral artery and femoral vein. Both catheters were tunnelled under the skin to exit through the scruff of the neck. The animals were awake and freely moving in a wooden box (20 cm × 12 cm × 11 cm). Arterial pressure was recorded via a Statham transducer P 23 Db and a Brush blood pressure computer on to a Brush 2400 polygraph. Changes in systolic blood pressure were evaluated. Heart rate was calculated from the pulse pressure wave by the computer. The intraventricular doses of enkephalins were: 0 (saline control), 0.36, 3.6, 36.0 and 360 nmol at an infusion rate of 2 μl/min. Each infusion lasted 10 min and after blood pressure had returned to baseline levels, the next infusion was started. The same doses of enkephalins (0, 0.36, 3.6, 36.0, 360 nmol) were injected intravenously as a bolus of 200 μl.

Four different groups of rats were studied: group 1 (n = 9) was treated with Leu-ENK; group 2 (n = 10) was treated with Leu-ENK and l-propranolol; group 3 (n = 9) was treated with Met-ENK; group 4 (n = 9) was treated with Met-ENK and propranolol. Groups 1–4 were tested on day 1 with intravenous and intraventricular enkephalins. Groups 2 and 4 then received an intraventricular l-propranolol infusion over 10 min (368 nmol), while groups 1 and 3 received intraventricular saline infusions. On day 2, the same doses of enkephalins were tested again in all groups of rats. In group 5 (n = 8), 772 nmol of l-propranolol was infused intraventricularly on day 1. On the next day, isoprenaline was injected intravenously at doses of 0.1, 1.0 and 10 nmol, and blood pressure recorded. Group 6 (n = 7): the rats were treated in the same way but no propranolol was administered.

Results are expressed as means ± SEM. Student’s t-test was used to evaluate the statistical significance of differences.

Results

Intraventricular infusions

Both Leu-ENK and Met-ENK produced similar dose-dependent increases of blood pressure (Fig. 1a) which were characterized by a slow onset and a minimum duration of 10 min. The doses of 0, 0.36, 3.6, 36.0, and 360 nmol of Leu-ENK resulted in respective increases in systolic blood pressure of 1.6 ± 1.2, 1.7 ± 1.1, 4.9 ± 2.0, 14.9 ± 4.3, 35.8 ± 6.3 mmHg. At the same doses Met-ENK produced increases of 0.5 ± 0.5, 0.2 ± 0.8, 5.5 ± 2.0, 22.3 ± 3.3, 37.3 ± 6.9 mmHg.

Heart rate also increased dose-dependently after intraventricular infusions of both Leu-ENK and Met-ENK (Fig. 1c). Leu-ENK resulted in respective increases in heart rate of 6.6 ± 7.4, 2.9 ± 9, 21.2 ± 8.9, 59.2 ± 19.8, 58.1 ± 24.9 beats/min. At the same doses Met-ENK produced increases of 1.5 ± 1.2, 2.1 ± 2.9, 18.3 ± 8.5, 54.4 ± 11.7, 77.1 ± 15.8 beats/min.

Intravenous injections

Leu-ENK and Met-ENK produced dose-dependent increases in blood pressure (Fig. 1b) which were characterized by a sharp onset and a rapid return to baseline levels within less than 1 min. The doses of 0, 0.36, 3.6, 36.0 and 360 nmol of Leu-ENK resulted in respective blood pressure rises of 0.4 ± 0.5, 0.3 ± 1.0, 2.7 ± 0.6, 20.2 ± 2.0 and 35.8 ± 2.7 mmHg. At the same doses Met-ENK produced responses of 0.2 ± 0.4, —3.7 ± 1.4, —0.8 ± 1.9, 25.8 ± 2.8 and 55.7 ± 3.6 mmHg. At the dose of 0.36 nmol, Met-ENK caused a fall in blood pressure, a change which was significantly different (P < 0.05) from that following Leu-ENK. At the dose of 360 nmol, the increase in blood pressure produced by Met-ENK was significantly (P < 0.01) greater than that produced by Leu-ENK.

Heart rate was not affected by intravenous
Blood pressure effects of enkephalins

Injections of Leu-ENK, whereas Met-ENK increased heart rate all doses given (Fig. 1d). The respective rise in heart rate were 1.4 ± 1.0, 16.6 ± 9.6, 17.2 ± 6.8, 22.7 ± 4.6 and 27.1 ± 11.3 beats/min.

Intraventricular infusions after intraventricular propranolol

Twenty-four hours after the infusion of 386 nmol of l-propranolol into the brain, the increases in blood pressure after both intraventricular Leu-ENK and Met-ENK were significantly \( P < 0.05 \) reduced at the highest dose of 360 nmol. At this dose, the increases in blood pressure before, versus those after, propranolol treatment were 39.8 ± 9.8 vs 19.0 ± 8.3 mmHg in the Leu-ENK group, and 35.3 ± 7.4 vs 13.5 ± 3.6 mmHg in the Met-ENK group.

The increases in heart rate after intraventricular Leu-ENK were not affected by intraventricular propranolol, whereas these following intraventricular Met-ENK decreased significantly \( P < 0.05 \) at the highest dose of 360 nmol. At this dose, heart rate increased by 90.0 ± 29.2 beats per min before, and by 19.2 ± 9.2 beats/min after propranolol.
Intravenous infusions after intraventricular propranolol

The responses of blood pressure and heart rate to intravenous Leu-ENK and Met-ENK were not affected by intraventricular propranolol.

Intravenous isoprenaline

The responses of blood pressure to intravenous isoprenaline were the same before and 1 day after propranolol treatment.

Discussion

Infusions of Met-ENK and Leu-ENK into the lateral ventricles of the brain and into the blood of rats led to marked increases in blood pressure and heart rate. These results are in accordance with those of other authors (Bisset et al., 1978; Bolme, Fuxe, Agnati, Bradley & Smythies, 1978). Similar increases in arterial blood pressure have also been obtained in conscious, freely moving cats after administration of β-endorphin and [D-Ala²]-ENK (unpublished work).

The type of blood pressure response following intravenous and intraventricular administration differed markedly. The increases in blood pressure following intraventricular infusions were characterized by a slow onset and a long duration of about 10 min, whereas a sharp onset and a rapid return to baseline levels within less than 1 min was observed after intravenous injections of Leu-ENK and Met-ENK. The different character of the blood pressure responses following intraventricular and intravenous administration was confirmed in control experiments with intravenous infusions instead of bolus injections and suggests different mechanisms of action in the periphery and in the brain. It also indicates that the intravenous effect is not brought about by stimulation of the same brain receptors, and vice versa that the intraventricular effect of enkephalin is not due to leakage of the peptide into the blood. In this respect, it is of interest that enkephalin was shown to release antidiuretic hormone and was associated with an antidiuretic response. This response was at least ten times more marked following intraventricular than intravenous administration (Bisset et al., 1978). The increases in blood pressure following intraventricular Leu-ENK and Met-ENK at the highest dose were reduced 24 h after intraventricular propranolol treatment. Only the intraventricular Met-ENK, but not the intraventricular Leu-ENK-induced increases in heart rate were reduced by propranolol. The responses to intravenous Leu-ENK and Met-ENK were not affected by intraventricular propranolol.

The different heart rate responses when intraventricular Leu-ENK and Met-ENK were given after propranolol, and also the lack of increase in heart rate following intravenous Leu-ENK, support the hypothesis that different types of receptors are involved in the cardiovascular effects of Leu-ENK and Met-ENK (Bolme et al., 1978). In contrast to morphin and other morphinomimetic peptides (Bolme et al., 1978), vasodepressor activity was only observed at low doses of Met-ENK. This may be explained by the injection site in these experiments, but may also be due to stimulation of different receptors. The relative resistance of the hypertensive effect to naloxone blockade (Bolme et al., 1978) supports this latter explanation. The blood pressure increases were independent of epileptic activity as shown by electrical recordings in cortical and subcortical brain areas of the cat (unpublished work).

The reduction by l-propranolol of the cardiovascular effects of enkephalin and angiotensin (Johnson et al., 1978) indicates that peptidergic transmitters or neurohormones can be influenced by β-adrenoceptor blockers, and that the sympathetic nervous system participates in their cardiovascular effects. The central action of l-propranolol has been verified by the lack of effect on the response to intravenous administration of isoprenaline in these experiments.

The results show that enkephalins may be involved in the haemodynamic adaptation which occurs under conditions of circulatory stress (Baker & Smith, 1977) through their central and peripheral effects on blood pressure and heart rate.

Acknowledgments

These studies were supported by the Deutsche Forschungsgemeinschaft within the SFB 90 ‘Cardiovaskuläres System’. The technical assistance of S. Thiele and the secretarial help of M. Funke is acknowledged.

References


Blood pressure effects of enkephalins


