Korean J. of Pharmacology
Vol. 24, No. 2, pp. 153-164, 1988

☐ Review Article ☐

Do Opioid Receptors Play a Role in Blood Pressure Regulation ?[1,2]

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ABSTRACT

The potential role of endogenous opioid peptides (EOPS) in cardiovascular regulation has only recently been entertained. EOPS have been localized in brain, spinal cord, autonomic ganglia, particularly the adrenal gland, and many other peripheral tissues. There are at least five major types of opioid receptors; namely μ, δ, k, σ, and ε. Experimental evidence indicates that cardiovascular actions of the peptide are mediated primarily by μ, δ and k receptors, and that these receptor types may be allosterically coupled. In anesthetized rabbits met-enkephalin decreased blood pressure and heart rate, which closely paralleled a reduction in sympathetic discharge.

Naloxone, but not naloxone methobromide, antagonized these effects, which suggests a central site of action of met-enkephalin. A number of autonomic agents, particularly adrenergic α- and β-agonists and antagonists modify the cardiovascular actions of met-enkephalin. Experiments in reserpine-treated and adrenalectomized rats provide no evidence of sympathetic nervous system involvement in the pressor responses to intravenous injection of opioid peptides, but rather suggest a direct peripheral action. Finally, activation of a beta-endorphinergic pathway projecting from the arcuate nucleus to the nucleus tractus solitarii in rats can cause naloxone reversible hypotension and bradycardia.

There is evidence to implicate this pathway in antihypertensive drug action and in the modulation of baroreflex activity.

Key Words: Opioid Peptide, Opioid Receptor, Cardiovascular, Blood pressure, Sympathetic System

INTRODUCTION

Tight regulation of systemic blood pressure is essential for life. Neurohumoral mechanisms, particularly the hypothalamus-pituitary-adrenal system, play an important role in the maintenance of blood pressure. Conventionally, this could be achieved by the release of adrenocorticotropic hormone from the anterior pituitary gland, of norepinephrine from sympathetic nerve terminals, and of epinephrine from the adrenal medulla (Axelrod *et al.*, 1984).

Recent experimental evidence has suggested that EOPS such as β-endorphin and enkephalins play an important role in cardiovascular regula-

[1]From the symposium "Endogenous Opioid Peptides and Regulation of Blood Pressure" sponsored by the American Society for Pharmacology and Experimental Therapeutics at its 38th Annual Meeting at Honolulu, Hawaii on August 15 to August 19, 1987.

[2]This symposium was supported in part by grants from E. I. De Pont de Nemours and Company, and The Upjohn Company.

tion (Holaday, 1983; Rhee *et al.*, 1985; Wightman *et al.*, 1987), in addition to their diverse physiological effects on pain perception, thermoregulation, obesity, endocrine systems, energy metabolism and pulmonary function (Holaday, 1985; Saprue *et al.*, 1981). Histological studies indicate that EOPS are widely distributed not only in the brain, but also in sympathetic nerves and ganglia (Schultzberg *et al.*, 1978). In particular, large amounts of enkephalins are present in the adrenal medulla of many mammalian species (Lewis *et al.*, 1980). From this site, co-secretion of catecholamines and enkephalins is induced under conditions of stress such as intense exercise or shock states (Chaminade *et al.*, 1983; Van Loon *et al.*, 1981).

In anesthetized animals, met-enkephalin administration leads to a reduction of blood pressure and heart rate by direct suppression of sympathetic discharge. There is ample evidence that EOPS interact specifically with catecholamines and other neurotransmitters which regulate systemic blood pressure. Thus, the primary objective of this symposium review is to recognize a functional role of EOPS and their receptors in the regulation of the cardiovascular system with a special emphasis on blood pressure regulation.

TYPES OF OPIATE RECEPTORS THAT MEDIATE CARDIOVASCULAR FUNCTION

Over the last few years, the physiological and pathophysiological functions of EOPS and their receptors have received increasing attention. There are several forms of opioid peptides, including those related to β-endorphin, enkephalins and dynorphins. Their actions are mediated by multiple opioid receptors including μ, δ, and k receptors (Table 1). Many of these peptides are distributed in nervous tissues, endocrine organs, as well as in the gastrointestinal system.

Due to the complexity of autonomic networks, the administration of opioid substances may produce different effects depending upon the site of injection, dose, and state of consciousness. These responses may reflect direct actions at cardiac or vascular sites or indirect actions through altered sympathetic outflow and/or through the release of vasoactive mediators (*e.g.* histamine). In conscious animals, the intravenous administration of opioids with predominantly μ actions (*e.g.* morphine at mg/kg doses) results in bradycardia and hypotension, whereas δ agonists (*e.g.* enkephalins at μg/kg doses) produce hypertension with little change in heart rate. The k agonist dynorphin (1 mg/kg) results in transient hypotension and little change in heart rate. However, these various agonist effects do not predict the physiological function of endogenous opioid systems.

The use of opioid antagonists have enabled investigations of the physiological and/or pathophysiological actions of EOPS. It has been shown that opioids inhibit baroreceptor reflexes by actions upon μ receptors. Studies defining the actions of opioid systems in various forms of shock and nervous system ischemia have revealed that different opioid peptides, and mechanisms of

Table 1. Opioid receptor subtypes and ligands

Receptor subtype	Endogenous agonists	Prototype agonists	Representative agonists	Representative antagonists
μ	β-Endorphin Met-enkephalin	Morphine	Morphiceptin Tyr-d-Ala-Gly-NMe-Phe-Gly-ol (DAGO)	Naloxonazine β-Funaltrexamine Naloxone β-chlornaltrexamine
δ	Leu-enkephalin	Leu-enkephalin	d-Ala^2-d-Leu^5-enkephalin (DADLE) d-Pen^2-d-Pen^5 enkephalin	ICI 154, 129 ICI 174, 864 16-Me-Cyprenorphine Naloxone
k	Dynorphin (1-17)	Ketocyclazocine	U 50 488 Dynorphin (1-17) Ethylketazocine	WIN 44 441-3 MR 2266 Naloxone
ε	β-Endorphin	β-Endorphin	β-Endorphin	Naloxone
σ	Unsequenced peptides	SKF 10,047 (N-allylnormetazocine)	Phencyclidine ?	?

action are involved. For example, endotoxic and anaphylactic shock involve predominantly δ-opioid receptors which affect sympathomedullary outflow and may alter catecholamine actions upon peripheral targets; hemorrhagic shock may affect both δ and μ receptors, which alter catecholamine responses at their sites of action in the heart (D' Amato *et al.*, 1984); and spinal injury and stroke may involve k receptors acting at local vascular sites within the nervous system.

Recent studies have revealed that these opiate receptor types may be allosterically coupled. Specifically, it was shown that μ antagonists or k agonists block the pressor effects of δ antagonists in endotoxic shock. Such interactions have been validated and extended using other models of in vivo and in vitro pharmacology. These data question the prior dogma that distinct opiate receptor types function independently. Instead, there may be a functional coupling among opiate receptor types as part of a large macromolecular complex.

MET-ENKEPHALIN, SYMPATHETIC NERVE ACTIVITY AND INTERACTION WITH ADRENERGIC AGENTS

In anesthetized rabbits, renal nerve activity represents primary sympathetic efferent activity (Fig. 1). Exogenous norepinephrine increased blood pressure which reflexly suppressed the renal nerve activity, whereas nitroprusside had the opposite effect. Met-enkephalin, norepinephrine or nitroprusside did not produce any significant changes in the afferent nerve discharge pattern (Fig. 1).

Met-enkephalin produced hypotension and bradycardia after an intravenous injection (Fig. 2). Since sympathetic nerve activity preceded the fall in blood pressure, the site of action of met-enkephalin appears to be central. In this animal

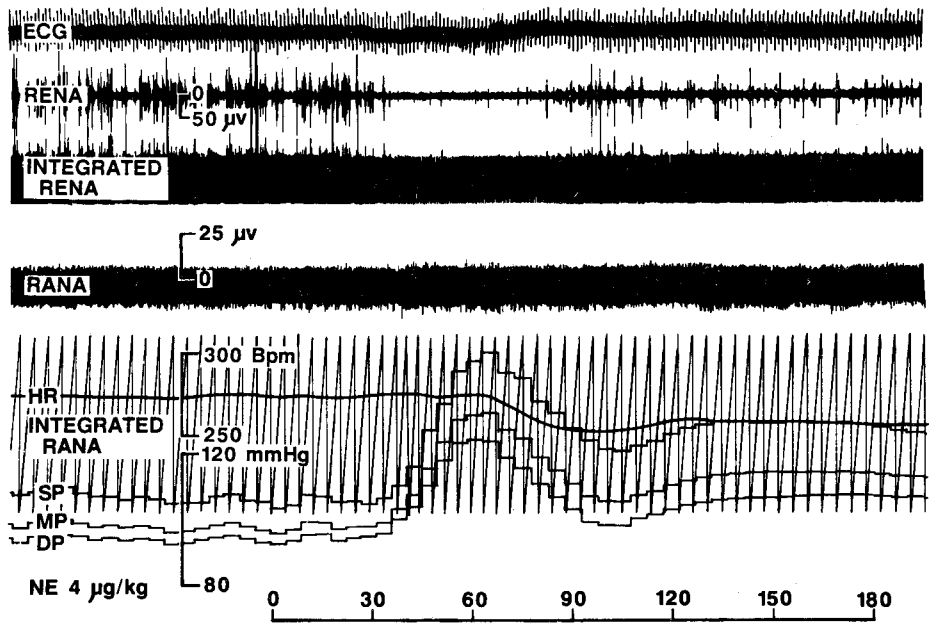

Fig. 1. Suppression of efferent renal sympathetic nerve activity by baroreflex mediated action of norepinephrine. In anesthetized rabbits norepinephrine (4 μg/kg) was injected intravenously at 0 second. Efferent renal nerve activity (RENA) was recorded from the proximal end of the nerve after it was severed.
Afferent renal nerve activity (RANA) was monitored from the distal end of the nerve. Integration of the nerve activities, recording of blood pressure and heart rate were carried out as reported (Rhee *et al.*, 1985). Sp, mp, dp and pp stand for systolic, mean, distolic and pulse pressures, which were obtained from a Gould pressure processor. Numbers at the bottom indicate time in seconds after the injection of norepinephrine.

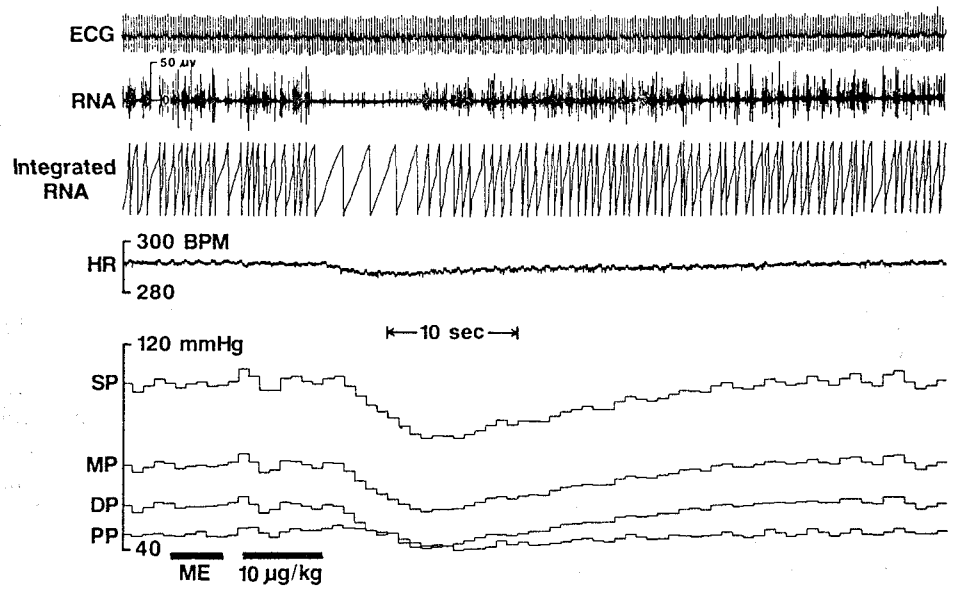

Fig. 2. Effects of met-enkephalin (ME) on sympathetic discharge, heart rate and blood pressure in anesthetized rabbits. In pentobarbital (30 mg/kg) anesthetized intact rats met-enkephalin (10 μg/kg) was loaded into a femoral venous catheter in a volume of 0.2 ml at the first black bar. The peptide was subsequently injected by pushing the catheter with 0.25 ml of saline at the second bar. All signs and symbols were used as in Fig. 1.

model, met-enkephalin also interferred with the baroreceptor-reflex, since the peptide reduced heart rate despite the fact that there was a significant reduction in blood pressure (Fig. 2). The hypotensive, bradycardiac and sympatho-inhibitory actions of met-enkephalin were blocked by pretreatment with naloxone (1 mg/kg), but not with naloxone methobromide, a quaternary analogue of naloxone (Rhee *et al.*, 1985). This confirms that the primary site of met-enkephalin action is within the central nervous system.

Although met-enkephalin decreased sympathetic nerve activity and subsequent blood pressure, the peptide had no effect on hypertension induced by exogenous norepinephrine (Eulie *et al.*, 1984). This supports again indirectly the central action of met-enkephalin, but a number of experimental evidence also suggests the idea that met-enkephalin acts directly on peripheral organs. Using radioactive microspheres, vascular resistance of 35 tissues of anesthetized rabbits was examined (Eulie *et al.*, 1987). Though there was a significant reduction in systemic blood pressure, vascular resistance in major muscles was significantly reduced (see Table 2). Activation of peripheral afferent systems by the treatment with met-enkephalin resulting in the reduction of sympathetic efferent activity is certainly a viable explanation. This view was supported by a recent study in hindquarters of conscious rabbits (Wightman *et al.*, 1987).

Pretreatment with prazosin (1 mg/kg, i.v.) or phentolamine blocks the cardiodepressive action of met-enkephalin. As phentolamine shows stronger inhibitory actions than prazosin, this suggests a presynaptic action of met-enkephaln. Indeed, clonidine, an adrenergic α_2 agonist, demonstrated an additive interaction with met-enkephalin in the suppression of renal nerve activity in anesthetized rabbits (Rhee, 1986).

Propranolol, but not its quaternary analog dimethyl propranolol, inhibited the bradycardic effect of enkephalin in anesthetized rabbits (Fig. 1) which suggests that the site(s) of interaction of propranolol and met-enkephalin is within the central nervous system. It is interesting that the bradycardiac, but not the hypotensive, action of propranolol (1 mg/kg) is antagonized by naloxone (1 mg/kg). In spontaneously hypertensive rats, however, naltrexone (2 mg/kg) antagonized the

Table 2. Antagonistic effects of naloxone on [Met^5] enkephalin induced vasodilation in the skeletal muscle of anesthetized rabbits[a]

Muscle	N[b]	Vascular resistance (mmHg · ml^{-1} · min^{-1} · per 100 g tissue)		
		Control	[Met^5] enkephalin alone[c]	[Met^5] enkephalin plus naloxone[d]
Right triceps	8	36.79±4.48	9.65±2.35[f]	39.85± 5.06
Left triceps	8	36.46±5.61	8.87±1.56[f]	42.42±13.62
Right pectoralis	8	22.29±5.18	9.06±1.98[e]	23.56± 5.15
Left pectoralis	8	24.11±6.23	8.77±2.05[e]	21.67± 5.24
Right masseter	9	22.19±4.08	13.43±3.11[e]	16.60± 3.66
Left masseter	9	20.15±3.14	9.12±2.10[e]	19.22± 2.71
Rectus abdomi	9	19.54±2.18	15.79±2.20	20.30± 4.93
Diaphragm	9	6.11±1.04	3.96±0.96	6.52± 1.28

[a] Vascular resistance was calculated by a formula of (pressur/flow) and all values are means±S.E. Mean arterial blood pressures in control, [Met^5] enkephalin and [Met^5] enkephalin after naloxone treatment were 85.3, 55.0 and 93.9 mmHg, respectively.

[b] Indicate number of animals used. [c] [Met^5] enkephalin dose was 1 mg/kg body weight.

[d] Naloxone (1 mg/kg) was administered 10 min before [Met^5] enkephalin. [e,f] Indicate P is less than 0.05 and 0.01 respectively, compared to the relevant control.

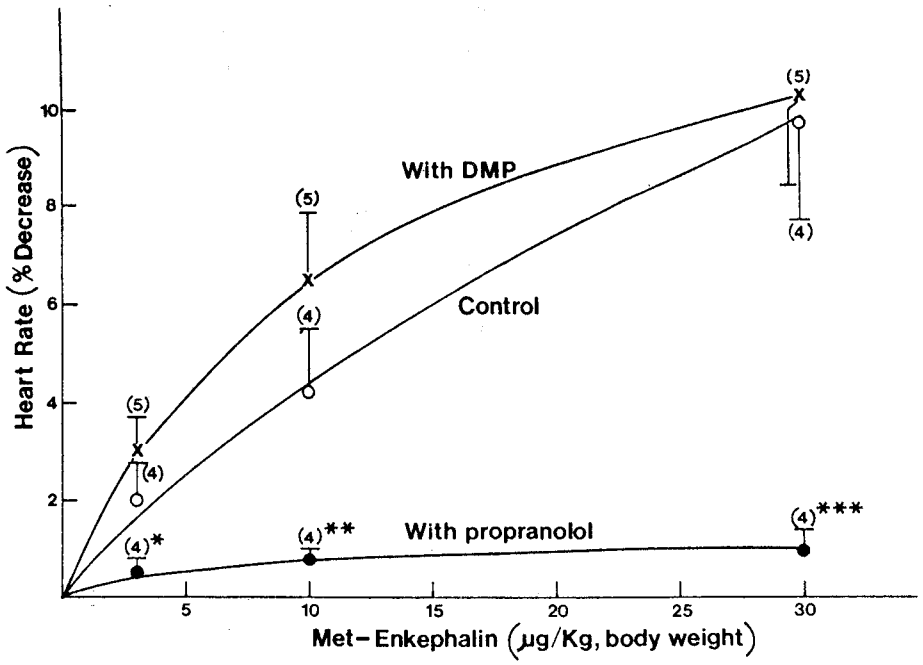

Fig. 3. Differential action of propranolol and dimethylpropranolol (DMP, UM272) on bradycardiac action of met-enkephalin. In anesthetized rabbit bradycardiac action of met-enkephalin was expressed as percent decrease from the codntrol heart rate after 3, 10 or 30 μg/kg of met-enkephalin. In different animals the same dose-response test was repeated 10 min after pretreatment with either i.v. propranolol (1 mg/kg) or i.v. dimethylpropranolol (1 mg/kg). Vertical bars indicate standard error and the numbers in the parentheses are the number of animals used. *, ** or *** indicate p is less than 0.05, 0.01, or 0.005, respectively.

depressor as well as bradycardiac actions of intracisternal propranolol (Ramirez *et al.*, 1982).

CHARACTERISTICS OF ADRENAL RELEASE OF EOPS AND CATECHOLAMINES

As indicated above, adrenal medullary chromaffin cells contain catecholamines and a variety of opioid peptides. The opioid pentapeptides met-and leu-enkephalin and their carboxy-terminal extended forms, released concurrently with catecholamines from chromaffin vesicles (Chaminade *et al.*, 1983) and from splanchnic nerve endings (Schultzberg *et al.*, 1978), may modulate the secretory adrenal medullary response via action on specific opioid receptors. Given that the secretion of these hormones constitutes the response of the sympathetic nervous system to stress and evidence that the hormones may be differentially released, an important question is, whether different secretory patterns are produced by different stress stimuli.

The concurrent adrenal release of met-enkephalin and catecholamines: norepinephrine, epinephrine, dopamine, evoked by different physiological stimuli was studied in halothane anesthetized cats. Adrenal venous blood was collected (300 μl/min) via a catheter in the left lumboadrenal vein under baseline conditions and during the electrical stimulation of the left splanchnic nerve or both sciatic nerves. Hemorrhage (50% of total blood volume) over 1-1.5 hrs. and splanchnic occlusion shock were also used as models of stimulation.

Table 3 represents the concurrent adrenal plasma levels of met-enkephalin and catecholamines during baseline conditions and their proportional changes under the different experi-

Table 3. Effects of stimulation on adrenal levels of met-enkephalin and catecholamines in anesthetized cats

Manipulation	Hemodynamics		Adrenal vein plasma levels				Molar ratios of adrenal vein hormones		
	BP (mmHg)	HR (bpm)	NE (ng/ml)	EPI (ng/ml)	DA (ng/ml)	ME (pg/ml)	NE/EPI	DA/EPI	ME/EPI $\times 10^{-3}$
Baseline: (n=20)	125 ±5	212 ±6	4.17 (3.16 5.50)	12.50 (8.17 19.13)	0.23 (0.17 0.31)	72.44 (57.49 91.27)	0.36 (0.28 0.46)	0.02 (0.01 0.03)	0.62 (0.42 0.92)
Stimulation effects:	Δ from Baseline		Post÷Pre-Stimulation Adrenal Plasma Levels				Post÷Pre-Stimulation Molar Ratio		
	BP	HR	NE	EPI	DA	ME	NE/EPI	DA/EPI	ME/EPI
Splanchnic Nerve: (n=10)	31* ±8	-25* ±11	51.29* (30.17 87.19)	40.74* (12.96 69.26)	11.48* (7.60 17.33)	4.68* (2.82 7.77)	1.25 (0.99 1.58)	0.28* (0.20 0.39)	0.11* (0.07 0.16)
Sciatic Nerve: (n=10)	21* ±4	3 ±9	13.18* (7.57 22.93)	6.92* (3.89 12.32)	8.91* (5.75 13.81)	2.00* (1.52 2.64)	1.86 (1.32 2.62)	1.26 (0.91 1.74)	0.29* (0.19 0.45)
50% Hemorrhage: (n=10)	-74* ±6	-3 ±11	112.20* (58.74 214.30)	34.67* (18.15 66.22)	60.26* (33.85 107.26)	10.47* (7.22 15.18)	3.24* (2.45 4.28)	1.74* (1.38 2.19)	0.46 (0.23 0.94)
Splanchnic Occlusion: (n=8)	-65* ±8	-13 ±14	131.83* (58.85 295.30)	43.65* (22.38 85.12)	190.55* (83.21 436.36)	19.50* (12.34 30.81)	3.02* (2.19 4.17)	4.37* (3.10 6.16)	0.45 (0.24 0.84)

Hemodynamic parameters, blood pressure (BP) and heart rate (HR), and adrenal vein plasma levels of norepinephrine (NE), epinephrine (EPI), dopamine (DA) and met-enkephalin (ME) in 20 cats under 1 MAC of halothane anesthesia under baseline conditions 1 hr after termination of the surgical procedure. Adrenal hormone levels were also converted to molar values and expressed as ratios of EPI. Effects of splanchnic nerve stimulation, bilateral sciatic nerve stimulation, 50% hemorrhage and splanchnic occlusion are expressed as change of individual group baseline levels for BP and HR, and as ratios of individual group baseline values for adrenal plasma levels of NE, EPI, DA, ME and as ratios of molar ratios at baseline for NE/EPI, DA/EPI and ME/EPI. Values are expressed as mean±SE for BP and HR, and as geometric means with limits of interval=geometric mean × ÷ geometric SE. (*$p<0.05$ compared to group base-line, paired T-test).

mental stimulations. While catecholamine concentrations in the adrenal vein are in the μg/ml range, met-enkephalin (pg/ml) on a molar basis only reaches concentrations 0.001 times than those of epinephrine. Direct stimulation of the splanchnic nerve evoked a significant increase in all catecholamines and met-enkephalin. Bilateral sciatic nerve stimulation evoked a significant rise in blood pressure and a different pattern of proportional hormone release. Again, the increase in met-enkephalin levels was significantly less compared to epinephrine. Slow hemorrhage of 50 % of total blood volume led to a significant fall in blood pressure and a prominent increase in adrenal catecholamines. The splanchnic occlusion shock model represented the strongest adrenal stimulus for all catecholamines and met-enkephalin at a time point when the blood pressure was significantly reduced from baseline.

Possible modulatory effects of opioids on the hemorrhage-evoked adrenal secretory response were evaluated by administration of specific opioid receptor agonists: sufentanil (25 μg/kg i. v.), metkephamid (3 mg/kg i.v.), U50488H (3.5 mg/kg i. v.) and the antagonist naltrexone (1 mg/kg i.v.) prior to induction of a 50% hemorrhage. None of these treatments led to a change in adrenal hormone levels as compared to a saline treatment group.

The relative levels of met-enkephalin and catecholamines in the adrenal plasma vary according to the applied stimulus. The dynamics of met-enkephalin release are less pronounced, approximately one-tenth as compared to epinephrine, suggesting a differential regulation of enkephalinergic and monoaminergic secretion. The apparently less marked decrease of molar ratios of met-enkephalin to epinephrine under 50% hemorrhage and splanchnic occlusion shock can be explained on the basis of marked increase in norepinephrine versus epinephrine levels, leading to a proportionally higher release of met-enkephalin from norepinephrine containing granules. The adrenal secretory response to bilateral sciatic nerve stimulation may be due to the effects of halothane anesthesia with a possibly selective suppression of pain-evoked adrenal sympathetic response. Baroreceptor-evoked adrenal sympathetic stimulation is neither modulated by opioid receptor agonists nor by an antagonist, thus excluding the involvement of opioids in the adrenal medullary response to hemorrhage in this in vivo model.

SIGNIFICANCE OF OPIOID PEPTIDE RESPONSE IN RESERPINIZED ANIMALS

Previously, Chandra and Dixon (1986) showed that D-als^2-methionine enkephalinamide gived intravenously in conscious, unrestrained rats increased mean arterial pressure. Naloxone antagonized this blood pressure increase when administered prior to D-ala^2-methionine enkephalinamide. In addition, intravenous injection of dynorphin (0.05 mg/kg to 0.4 mg/kg) also produced a blood pressure increase (Fig. 4). However, the pressor response to dynorphin was not affected by naloxone. In reserpine treated rats, the pressor response to D-ala^2-methionine enkephalinamide was similar but the duration of the pressor response was significantly longer compared to the response in rats not treated with reserpine. However, the pressor response to dynorphin was potentiated, while the duration of the pressor response was unchanged in rats treated with reserpine (Fig. 4).

The heart rate of normal rats was decreased significantly by intravenous injections of D-ala^2-methionine enkephalin or dynorphin (Fig. 5), but the decrease was much greater after D-ala^2-methionine enkephalinamide administration. In reserpine-treated rats, both D-ala^2-methionine enkephalinamide and dynorphin produced blood pressure increases accompanied by a decrease in heart rate (Fig. 4, 5).

In conscious, unrestrained rats the effects of intravenous administration of opiate peptides may be a result of complex interaction of the opiates with adrenergic and cholinergic systems within the myocardium. The finding that atropine given prior to the opiate peptides prevents the bradycardia is consistent with an interaction of opiates with muscarinic receptors in the myocardium. Therefore, variable effects of intravenously administered opiates on heart rate may be due to different basal activity of the vagal neuronal system within the myocardium resulting in varying degrees of vagal activation by the administered opiates. Although the blood pressure changes seen after intravenous administration of opiates is not directly related to opioid interactions with muscarinic receptors within the myocardium, a precipitous fall in the heart rate could mask any effects of the opioids to increase blood pressure by a direct effect on the

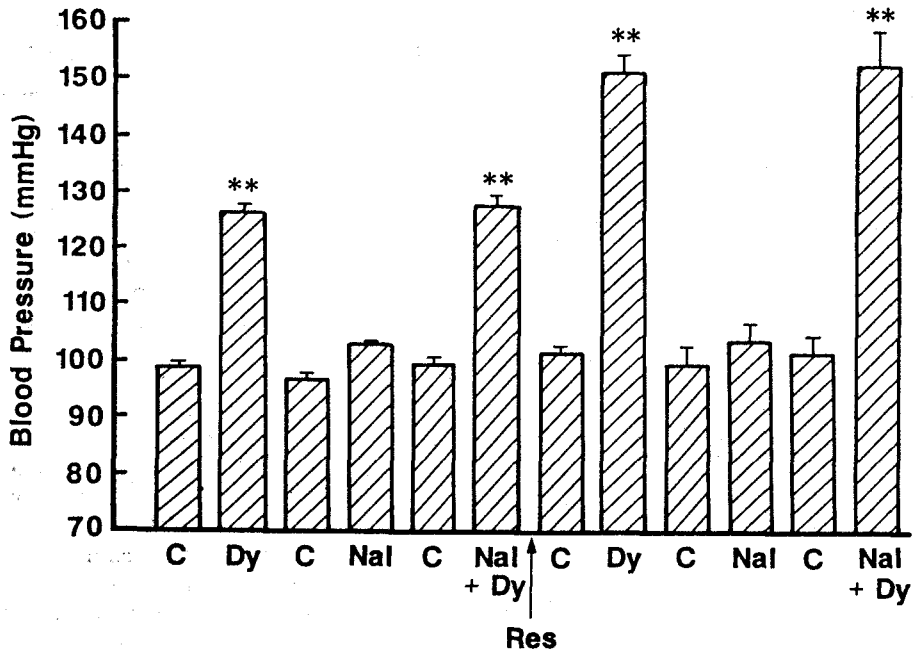

Fig. 4. Effect of dynorphin on blood pressure responses of conscious, unrestrained, normal, and reserpine-treated rats. The blood pressure was recorded continuously via a cannula in the femoral artery, and the blood pressure response to intravenous injections of dynorphin and naloxone in normal and reserpine-treated rats was determined. Statistical evaluation of the data was performed by ANOVA and by he Student Newman-Keul test for multiple comparisons of means. C indicates Control; Dy for dynorphin (0.1 mg/kg, i.v.); Res for reserpine (1 mg/kg, i.p.); Nal for naloxone (5 mg/kg, *i.v.*), respectively. **$p<0.01$ Dynorphin pressor response greater than the previous control, and **$p<0.001$ Dynorphin pressor response of reserpine-treated rats greater than dynorphin pressor response in control animals.

vasculature. This mechanism could, in part, be responsible for the varying effects of opioid peptides on blood pressure reported in the literature.

The results in reserpinized and adrenalectomized rats provide no evidence for sympathetic nervous system involvement in the pressor responses after intravenous injection of opiates. In fact, the pressor response to dynorphin was potentiated while the duration of the pressure response to D-ala^2-methionine enkephalinamide was prolonged in reserpinized rats in comparison to control rats. Therefore, it is likely that the pressor response to intravenously administered D-ala^2-methionine enkephalinamide and dynorphin results from a direct action on the vasculature. It is of interest that dynorphin was effective in raising the blood pressure at a much lower dose (one-tenth) than D-ala^2-methionine enkephalinamide and that naloxone antagonized the pressor effects of D-ala^2-methionine enkephalinamide but not dynorphin.

THE ROLE OF β-ENDORPHIN IN CARDIOVASCULAR REGULATION

Studies from several laboratories have demonstrated that the hypotensive and bradycardiac effects of clonidine and α-methylnorepinephrine, drugs acting at central α_2-adrenergic receptors, have naloxone-reversible components in rats (Farsang *et al.*, 1979; Kunos *et al.*, 1987; Petty *et al.*, 1984). Since clonidine and naloxone do not cross-react with each other's specific binding sites in the brain (Farsang *et al.*, 1979), their interaction has been taken to indicate the involvement of an endogenous opioid in the effects of clonidine-like agents.

This opioid appears to be β-endorphin, for several reasons: ⓐ β-endorphin administered cen-

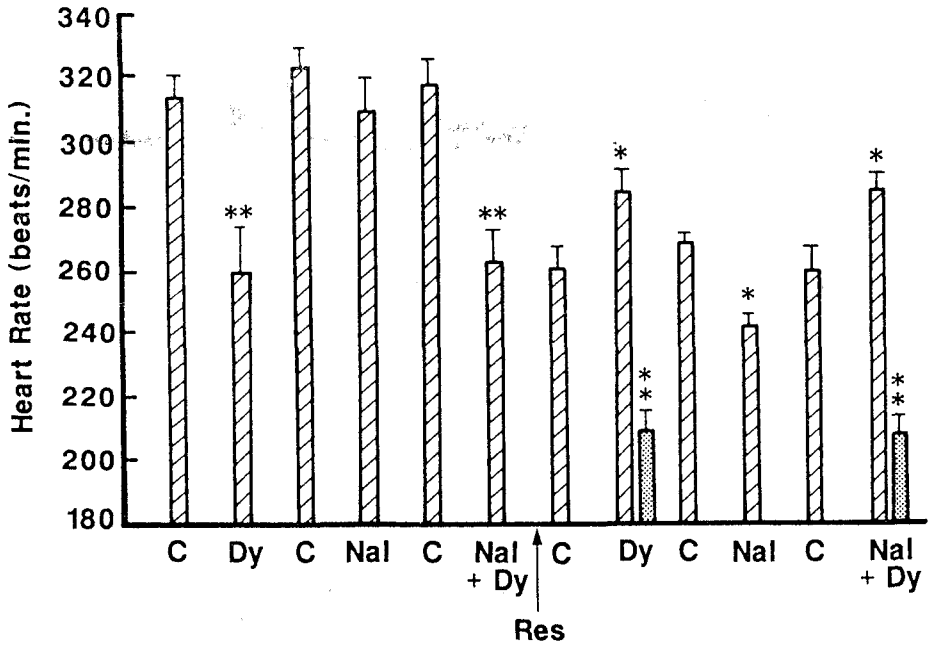

Fig. 5. Effect of dynorphin on heart rate response of conscious, unrestrained, normal and reserpine-treated rats. The heart rate was recorded continously via a cannula in the femoral artery and the heart rate response to intravenous injections of dynorphin and naloxone in normal and reserpine-treated rats was determined. Statistical analysis and all signs were used as in Fig. 4. The heart rate response to dynorphin was biphasic in reserpine-treated animals, a transient increase followed by a decrease as indicated by clear bar and black bar, respectively.

*$p<0.05$ significantly greater or less than previous control.

**$p<0.01$ significantly less than previous control, and

‡$p<0.01$ significantly less than previous control.

trally or systemically causes hypotension and bradycardia; ⓑ clonidine-like agents release immunoreactive β-endorphin from brain stem in vitro; and ⓒ central administration of antisera to β-endorphin antagonizes the cardiovascular effects of clonidine-like agents (Kunos *et al.*, 1987). The most likely site of action of the released β-endorphin is the brainstem nucleus of the solitary tract (NTS). An intra-NTS injection of 100 ng of naloxone stereo-selectively inhibited the effects of clonidine in both normotensive Sprague-Dawley and spontaneously hypertensive rats (Kunos *et al.*, 1987; Mosqueda *et al.*, 1986). These findings do not indicate, however, the source of the β-endorphin released by clonidine.

Neonatal treatment of rodents with monosodium glutamate is known to selectively destroy endorphinergic neurons of the brain that originate in the arcuate nucleus, without affecting β-endorphin in the pituitary. In spontaneously hypertensive rats, such treatment was shown to abolish the naloxone-sensitive component of the cardiovascular effects of clonidine (Mosqueda *et al.*, 1986), and electrolytic lesions to the mediobasal hypothalamus were found to have a similar effect (Mastrianni *et al.*, 1987). This suggests that the source of the β-endorphin involved in the actions of clonidine is in axon terminals in the NTS, which originate in the arcuate nucleus.

The possible cardiovascular regulatory role of neuronal pathways originating in the arcuate nucleus was further investigated in normotensive rats anesthetized with urethane. A bipolar electrode was stereotaxically inserted into the left arcuate, using the coordinates AP: −2.5 mm, ML: 0.1 mm and DV: −10.1 mm from the bregma. The position of the electrode was verified by postmortem examination. Blood pressure (BP) was monitored through an intraarterial cannula connected to a pressure transducer and polygraph.

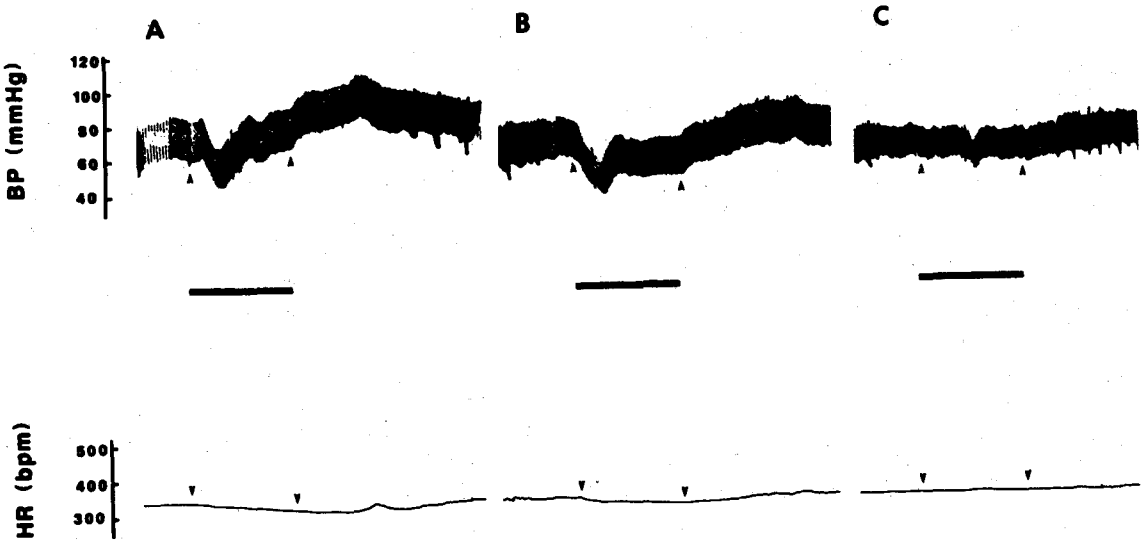

Fig. 6. Cardiovascular effects of electrical stimulation of the arcuate nucleus in a urethane-anesthetized Sprague-Dawley rat. A male rat weighing 300 g was anesthetized with urethane, 0.8 g/kg i.v.+0.3 g/kg i.p. Details for placement of the stimulating electrode and the measurement of BP and HR are given in the text. Electrical stimulation was at 250 μA, 0.8 ms, and 20 Hz, for a period of 30 sec, as indicated by the horizontal lines.

The beginning and end of stimulation is also marked by small arrows. Original recording of phasic BP (top) and HR (bottom) is shown. A: control stimulation; B: stimulation 10 min after the i.v. injection of 10 μg/kg of [1-(β-mercapto-β, β-cyclo-pentamethylenepropionic acid, 2-(0-methyl) tyrosine] arginine vasopressin; C: stimulation 20 min after the i.v. injection of 2 mg/kg naltrexone HCl.

Heart rate (HR) was monitored through a tachograph preamplifier, driven by the pressure wave. Electrical stimulation of the arcuate caused hypotensive or, more commonly, biphasic, depressor/pressor responses.

In any one animal, the control response was very reproducible upon repeated stimulations at 30 min intervals. As illustrated by a typical experiment, the stimulation-induced initial decrease in BP was gradually reversed by an increase in BP, which peaked only after stimulation had been terminated, while the bradycardiac response was maintained during the stimulation (Fig. 6A). When stimulation was repeated 10 min after the i. v. administration of a vasopressin V_1-antagonist, the pressor component was blunted, resulting in unopposed hypotension and bradycardia (Fig. 6B). Subsequent administration of naltrexone, 2 mg/kg i.v., blocked both the hypotensive and bradycardic responses (Fig. 6C).

These findings suggest that an endorphinergic pathway originating in the arcuate nucleus can activate the efferent baroreflex arc and may serve to facilitate the depressor baroreflex response (Ramirez *et al.*, 1983). Additional projections from the arcuate may be involved in vasopressin-mediated pressor mechanisms. These latter projections may be to the paraventricular nucleus, as suggested recently by others (Brody *et al.*, 1986). The present findings implicate brainstem endorphinergic neurons not only in antihypertensive drug action, but also in the modulation of baroreflex activity.

CONCLUSION

There is little doubt about the significant role of EOPS in the regulation of blood pressure in mammals. In this symposium review, the emphasis was placed on the effects of opioid peptides on central sympathetic discharge and on the adrenal release of the peptides and other neurotransmitters. Central sites, particularly at the level of the brainstem, are likely the main site(s) for the cardiovascular depressor actions of EOPS, while the pressor

responses to EOPS may be due either to central actions, such as the suppression of the baroreflex, or to actions at peripheral opioid receptors as presented in experiments with reserpinized animals.

As outlined, the cardiovascular actions of EOPS are the final manifestations of complicated interactions between the central and peripheral components of neural and humoral systems. Further studies on the effects of opioid peptides on synthesis and metabolism of major neurotransmitters, particularly on cellular fluxes of mono-and di-valent cations are needed for the elucidation of the cellular mechanism of opioid peptides.

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