

# The opioid system in cardiac and vascular regulation of normal and hypertensive states

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**ABSTRACT** The endogenous opioid system includes three major families of peptides: dynorphins (derived from pre-proenkephalin B), endorphins (derived from pre-proopiomelanocortin), and enkephalins (derived from pre-proenkephalin A). Multiple species of opioid peptides are derived from these major precursors and many of them possess potent cardiovascular properties. Opioid peptides and opioid receptors, of which multiple forms have been defined, are present in the central nervous system and peripheral neural elements. In the central nervous system, opioid peptides and receptors are found in forebrain and hindbrain nuclei involved in baroregulation, sympathoadrenal activation, and several other vital autonomic functions. In the periphery, opioid peptides are found in autonomic ganglia, adrenal gland, heart, and other organs; multiple opioid receptors are also found in vascular tissue, heart, and kidneys. Although little is known to date on the regulatory mechanisms of the opioid system in normal cardiovascular states, it became clear that cardiovascular stress situations substantially modify the activity of the endogenous opioid system. The purpose of this review is to clarify the sites of interaction of the opioid system with all major components of the cardiovascular system and indicate the potential role of this system in the ontogenesis of cardiac malfunction, vascular diseases, and hypertension.

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OPIATES are among the oldest pharmacologic substances known to man. Besides analgesia, one of the earlier (1824) observations on the biological activity of morphine and other opiate alkaloids was the demonstration of potent cardiorespiratory effects of these agents.<sup>1-4</sup> The cardiac, respiratory, and blood pressure effects of morphine were reported by several authors in the second half of the 19th century.<sup>3, 5, 6</sup> Since the discovery of opioid peptides and the identification of multiple opioid receptors in the heart and brain nuclei involved in cardiorespiratory control,<sup>7, 8</sup> the role of opioid peptides in autonomic regulation of blood pressure, heart function, and respiration became even more complex. Endogenous opioid peptides are a heterogeneous group of peptides, which in spite of some important structural similarities differ from each other in respect to synthesis, distribution, release, and actions.<sup>9</sup> Their actions are mediated by a family of receptors that may recognize different opiates and opioid peptides in different ways to yield variable, sometimes opposing, cardiovascular effects. The presence or absence of an-

esthetics can drastically change opiate effects. The aim of this review is to focus on the conflicting experimental data on the cardiovascular actions of opioid peptides and to summarize the present knowledge on the role of the opioid system in cardiovascular control and their possible significance in the pathophysiology of hypertension.

**Site and mode of the cardiovascular actions of opiates and opioid peptides.** Opioid peptides and their receptors are present in brain areas important for cardiovascular control, in heart, in autonomic ganglia, and in adrenal medulla.<sup>7, 9-12</sup> Exogenous opioid peptides/opiates induce different effects on blood pressure and heart rate depending on the route of administration or on anesthesia. Pressor responses and tachycardia follow the lateral brain ventricular administration,<sup>13</sup> but hypotension and bradycardia are observed after systemic injections of morphine or opioid peptides.<sup>13, 14</sup> The differences in effects of opioids after systemic vs central administration are often used to indicate lack of penetration in sufficient quantities to the cardiovascular sites in the brain. However, the controversy between these routes of administration can be the result of opposite effects of the peptide in different brain sites, resulting in summation response with little or no effect on the gross cardiovascular variables. For example, Feuerstein and Faden<sup>15</sup> found pressor and depressor sites for morphine and D-Ala<sup>2</sup>,D-Leu<sup>5</sup>-enkephalin (DADL) in neighboring hypothalamic nuclei, which were less than 1 mm apart from each other. Opposite blood pressure responses can be recorded also after injections of various

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enkephalin analogs into the anterior hypothalamic area of pentobarbitone-anesthetized rats vs conscious animals: hypotension accompanied by bradycardia in the anesthetized vs increases in blood pressure and heart rate in the conscious rat.<sup>16-18</sup> Since the discovery of multiple opiate receptors in the brain,<sup>12, 19, 20</sup> new data on the specific effects mediated by different receptors have accumulated. Development of new selective agonists and antagonists for the appropriate opioid receptors has made it possible to study the effects of different opioid receptor stimulation in different brain nuclei. For example, D-Ala<sup>2</sup>-MePhe<sup>4</sup>-Gly<sup>5</sup>-ol-enkephalin (DAGO) and morphiceptin are highly selective ligands for the mu-receptor,<sup>21, 22</sup> DADL a relative specific agonist for delta-receptor,<sup>23</sup> and dynorphin A and the benzomorphan derivatives such as bremazocine are considered to act mainly on the kappa opioid receptor.<sup>24, 25</sup> By means of stereotactic microinjections into the discrete brain nuclei associated with cardiovascular control, it has been shown that DAGO is about 100-fold more potent than DADL in inducing hypotension and bradycardia upon injections into the anterior hypothalamus of pentobarbitone-anesthetized rats.<sup>16</sup> Similarly, DAGO microinjected into the nucleus tractus solitarius is over 10-fold more potent than DADL in eliciting increases in blood pressure and heart rate in the spontaneously breathing or artificially respiration rat.<sup>26</sup> Qualitative differences in the cardiovascular effects of various opiate agonists can also be observed when kappa-receptor agonists are compared with mu/delta agonists.<sup>27</sup>

The interference of the opioid peptides with the autonomic nervous pathways suggests involvement of both the sympathetic and parasympathetic nervous systems in the cardiovascular actions of these peptides. An activation of the sympathoadrenomedullary axis seems to underly the pressor and tachycardiac responses to the mu-agonist DAGO or the delta-agonist DADL in the conscious rat.<sup>18, 28, 29</sup> The bradycardic effects of various enkephalin analogs, on the other hand, have been attributed to an activation of the vagus,<sup>18, 30, 31</sup> although the potent respiratory depressant effect may be the main determinant of this effect.<sup>16-18, 27</sup> The plasma vasopressin levels have been shown to be depressed after intracerebroventricular<sup>32, 33</sup> and intrahypothalamic injections<sup>28</sup> of opioid peptides, but no correlation between the effect of opioids on vasopressin on the one hand and blood pressure on the other hand have been demonstrated. In conclusion, the cardiovascular effects of opioid peptides seem to be mediated mainly by activation of the sympathetic nervous system, although a vagal component is likely to be

involved in some of their effects, whereas the release of vasopressin seems not to be important to the cardiovascular actions of opioid peptides.

**The opioid system in hypertension.** Hypertension is associated with significant changes in the opioid system. For example, the levels of dynorphins and leu-enkephalin in the pituitary gland and several brain nuclei are substantially different in spontaneously hypertensive rats (SHR) as compared with the normotensive animals.<sup>34</sup> Alterations in the brain opioid receptors in both experimental and genetic hypertension have also been reported.<sup>35-37</sup> The opioid system has been implicated in the development of high blood pressure in SHR, since the pressor responses to intracerebroventricularly administered opioid peptides in SHR are enhanced compared with those in normotensive rats.<sup>32, 33, 38, 39</sup> The partial opioid antagonist diprenorphine was also shown to produce a more pronounced hypotensive response in SHR than in normotensive Wistar-Kyoto rats (WKY),<sup>39</sup> whereas naloxone failed to affect blood pressure in either renal or genetic hypertensive rats.<sup>35</sup> Recently, it was shown in our laboratory that SHR differ also from normotensive rats in their response to hypothalamic administration of enkephalins; the SHR showed an augmented pressor response to the mu-agonist DAGO or the delta-agonist DADL microinjected into the medial preoptic nucleus of hypothalamus. These enkephalin analogs had little effect on heart rate of SHR, whereas in normotensive WKY a pronounced tachycardia accompanied the pressor response.<sup>28</sup> Again, the selective mu-agonist was more potent in the pressor effect than the delta-agonist.

The antihypertensive drug clonidine was reported to increase  $\beta$ -endorphin release from the brain slices of SHR,<sup>40</sup> whereas opiate antagonists blunt the hypotensive effect of clonidine.<sup>41, 42</sup> However, contradicting findings have recently been reported by Conway et al.<sup>43</sup> and Head and de Jong,<sup>44</sup> who found no evidence for involvement of endogenous opioid peptides in the effects of clonidine on blood pressure or plasma catecholamines. Since clonidine (an antihypertensive agent) lowers blood pressure, and exogenous opioid peptides can induce both hypertensive and hypotensive effects, these findings might further indicate a role of the endogenous opioid system in the development and/or maintenance of essential hypertension.

**Cardiac effects of opiates.** The heart is known to contain both opioid peptides and opioid receptors. The enkephalins found in the heart are located in nerve endings.<sup>45</sup> The enkephalins<sup>46</sup> were shown to decrease the chronotropic response of isolated spontaneously

beating rat atria to adrenergic agonists. The reduction in catecholamine-induced tachycardia was related to inhibition of  $\text{Ca}^{++}$  accumulation by the rat atria.<sup>47</sup> In the spontaneously beating guinea pig atria, however, leu-enkephalin augmented the chronotropic response to norepinephrine.<sup>48</sup> The inotropic response to norepinephrine in this preparation was not affected by either naloxone (a potent opiate antagonist) or by leu-enkephalin. Interestingly, the augmentation by leu-enkephalin of the chronotropic response to norepinephrine was also accompanied by a naloxone reversible increment in  $\text{Ca}^{++}$  uptake by atrial preparation.<sup>48</sup> Although the above cited data provide conflicting results on the nature of enkephalin modulation of cardiac rhythm by catecholamines (which might well represent species differences), they clearly show that opioid peptides, through activation of opioid receptors, have a primary action on the heart.

These pharmacologic studies are supported by biochemical data showing the presence of opiate receptors in the heart. Such evidence has been raised for the first time by Simantov et al.,<sup>49</sup> who detected specific binding of dihydromorphine and naloxone to membranes from whole hearts of rats and guinea pigs. However, in these original studies only very low levels of specific binding could be shown (5%). These studies were further supported by the results of Burnie,<sup>50</sup> who used a rat right ventricular preparation and diprenorphine as a nonselective opiate ligand. In none of these studies was the type of the opioid receptor in the heart characterized.

More recently, we have shown that most of the specific binding of diprenorphine to cardiac muscle membranes is found in the atria, in which receptor density is almost twice that in the right ventricle and threefold over the binding found in the left ventricle.<sup>7</sup> Characterization of these binding sites by sequential displacement of diprenorphine by selective ligands to the delta-receptor (DADL), the mu-receptor (DAGO), or the kappa-receptor (ethylketocyclazocine) strongly suggested the presence of delta- and kappa- but not the mu-receptor in the right atrium or ventricle. Interestingly, the binding of diprenorphine to membrane preparations from the right atrium revealed unusual cooperative binding.<sup>7</sup>

Furthermore, the opioid receptors found in the right ventricle and the right atrium can be down-regulated (by -45% and -60%, respectively) after acute bleeding (hemorrhage); the number of opiate receptors in the heart remains low as much as 24 hr after the bleeding. This phenomenon is opposite to the effect of hemorrhage on opiate receptor number in the brain, when up-

regulation of delta- and kappa-binding sites are apparent in the brain stem of rats exposed to hypovolemic hypotension.<sup>51</sup> The difference between the central and peripheral opiate receptor response to hemorrhage might be the result of excessive enkephalins/endorphins in the circulation after bleeding<sup>52</sup> and down-regulation phenomenon as a result of excessive agonist.

The significance of the presence of kappa-receptors in the heart is less clear, since circulating levels of dynorphin A (the putative endogenous kappa-receptor ligand) has not been determined as yet. However, intravenous administration of the kappa-agonist ethylketocyclazocine to dogs<sup>53</sup> or rats<sup>54</sup> can slow the heart rate.

These data taken together further support a role for selective opioid peptides and receptors in regulation of cardiac functions.

**Vascular tissue and the opioid system.** The role of the opioid system in regulation of vascular smooth muscle tone is poorly investigated. Yet, various opioid peptides are found in the circulation and elevated levels of opioid peptides in the plasma are found during cardiovascular stress situations (e.g., hypovolemia). The opioid peptides in the circulation might be derived from the pituitary gland ( $\beta$ -endorphin), the adrenal gland (enkephalins), or even peripheral organs such as the heart (dynorphin).

In vitro, opiate compounds like morphine, cyclazocine, and others have been shown to contract rat aortic strips.<sup>55</sup> The contractions produced in aortic strips by the opiates were highly dependent on  $\text{Ca}^{++}$  and were blocked by naloxone and verapamil (calcium entry blocker). These studies were substantiated later by others, showing that natural and synthetic opiates can contract rat aortic strips; however, the latter studies also showed that other blood vessels from the same species respond with profound relaxation (mesenteric arterioles), whereas muscular venules do not respond to the narcotic agents.<sup>56</sup> These authors have also shown that naloxone or  $\text{H}_1$  (histamine) receptors can prevent the narcotic induced vasodilation of the rat mesenteric arterioles, indicating an indirect mechanism (histamine dependent) for the narcotic effect on some blood vessels. Nevertheless, these pharmacologic studies strongly suggest the presence of opiate receptors in the vascular smooth muscle. Further studies on the cellular mechanisms of opiate interaction with vascular smooth muscle indicate that morphine-induced relaxation of the cat middle cerebral artery is the result of hyperpolarization of the muscle cells, which enhances relaxation.<sup>57</sup> This phenomenon was blocked by naloxone, indicating that opiate receptors are involved in the electromechanical effects of morphine on the smooth

muscle cell. These latter studies have also suggested a role of opiates in regulation of K<sup>+</sup> conductance in vascular smooth muscle.

Yet some effects of the opioids on smooth muscle tone might be mediated through naloxone-insensitive receptors. Thus, pulmonary vasoconstriction provoked by leu<sup>5</sup>-enkephalin in isolated rat lungs is not blocked by naloxone.<sup>58</sup> This finding was further substantiated by the demonstration of the lack of an effect of other opiates and opioid peptides (e.g., morphine or met<sup>5</sup>-enkephalin) in this same smooth muscle preparation. Moreover, the lack of effect of diprenorphine or DADL on this preparation was argued to exclude delta-opiate receptors as mediator of the leu<sup>5</sup>-enkephalin effect on the rat pulmonary artery.<sup>59</sup> However, in a different species, the rabbit, opiate-induced inhibition of norepinephrine release from the ear artery was argued to be predominantly mediated through delta-opiate receptors.<sup>60</sup>

The nature of the opioid effects on vascular smooth muscle is further complicated by the demonstrations that the opioid peptide dynorphin A(1-13), a preferential endogenous ligand for the kappa-opiate receptors, inhibits vascular smooth muscle contractions produced by electrical stimulation; in this regard, dynorphin A(1-13) is more potent than mu- or delta-opioid agonists.<sup>61</sup>

In summary, it is apparent that opiates and opioid peptides have direct effects on vascular smooth muscle tone. However, it is also clear that some opiate effects on vascular smooth muscle might not be direct (e.g., histamine mediated) or may not involve a naloxone-sensitive opiate receptor. It is also clear that the nature of the opioid effects on vascular tone varies according to the species and even in the same species, between different vascular beds. Mu-, delta-, and the kappa-opiate receptors seem to have some role in the different species and vascular preparations tested so far. The potential role of the opioids/opioid receptors in vascular tone regulation in hypertension remains to be elucidated.

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