Opioid physiology

Biosynthesis of morphine in the animal kingdom

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Two years ago, I wrote a News and Views article¹ entitled "Has morphine a physiological function in the animal kingdom?". New data, reported by Charles Weitz, Kym Faull and Avram Goldstein in their paper on page 674 of this issue², provide evidence that may answer this question.

When I wrote my previous article, it was generally considered that the endogenous opioids in animal tissues are fragments of the large peptide precursors pro-opioid, pro-enkephalin and prodynorphin, whereas the non-peptide morphine is found in plants such as the poppy (*Papaver somniferum*). But about three years ago, Spector and colleagues showed³ that immunoreactive morphine is present in low concentrations in toad skin (3 pmol g⁻¹ tissue); they found other non-peptide opioids in lower concentrations in the skin of rat and rabbit and, particularly, in bovine adrenal gland and brain (0.05–0.13 pmol g⁻¹). Shortly afterwards, Goldstein and his group identified⁴ immunoreactive morphine in bovine hypothalamus and adrenal gland. They stressed that it was not clear whether the very small amount of morphine they had found was of endogenous or exogenous origin. Therefore, demonstration of biosynthesis of morphine would be decisive. This consideration is of importance because, in 1981, Hazum *et al.* found⁵ immunoreactive morphine in cow's milk and suggested a possible dietary source in animal fodder.

Last year, Spector's and Goldstein's groups both presented the results of their further analyses of endogenous non-peptide opioids. Goldstein and his colleagues found⁶ that morphine and codeine are consistently present in extracts of bovine hypothalamus but variable in extracts of bovine adrenal gland and rat brain. Even then they still could not decide whether the action of these two morphinans is of an endogenous or exogenous nature or whether the morphinans are involved in brain function.

A few months earlier, Spector and

colleagues had found⁷ free codeine and free morphine in extracts of rat brain. Because in the morphine biosynthesis of *P. somniferum*, norlaudanosoline, reticuline, salutaridine, thebaine and codeine are precursors of morphine⁸, Spector and colleagues decided to test whether intravenous injection of these morphinans would increase the free morphine content in the brain and other tissues of the rat (Table 1). It was not known whether the concentration of free morphine obtained by intravenous injection of codeine (16

vitro. This critical step of conversion is not present in rat brain or bovine adrenal gland. The authors point out that this finding is the first demonstration of the ability of an animal tissue to synthesize morphinans. This result strongly supports the hypothesis that the codeine and morphine found in mammalian tissue are of endogenous origin.

Although the concentrations in animal tissues of free endogenous codeine and morphine are much lower than the concentrations of opioid peptides (Table 2), some speculation on their significance may be permissible. There are two significant differences between the peptide and non-peptide compounds^{10–12}. First, morphine interacts almost exclusively with the μ -opioid receptor, having a relative affinity of 0.98 (maximum = 1.0; Table 2) but [Met]enkephalyl-Arg-Arg-Val-NH₂ is less selective, having a relative affinity of 0.66

Table 2 Peptide and non-peptide opioid compounds with preference for the μ -site

Compounds	Relative selectivity for μ -binding in guinea-pig-brain ¹¹	Content (pmol g ⁻¹)	Tissue
[Met]enkephalyl-Arg-Arg-Val-NH ₂	0.66	6.5	bovine hypothalamus ¹²
[Met]enkephalyl-Arg-Phe	0.60	112	bovine hypothalamus ¹²
Morphine	0.98	0.03 (free) 0.7 (total)	rat brain ^{7,9}

Selectivity index: K_i^{-1} for $\mu/(K_i^{-1}$ for $\mu + K_i^{-1}$ for $\delta + K_i^{-1}$ for κ).

pmol g⁻¹; Table 1) is high enough to exert an agonist effect on the μ -opioid receptor. Spector and his colleagues have now shown⁹ that free morphine and codeine are only small fractions of the sum of free and sulphate-conjugated compounds. In the rat brain, free morphine is 0.03 pmol g⁻¹, whereas the mean of total morphine is 0.7 pmol g⁻¹ and that of total codeine is 1.2 pmol g⁻¹.

These data indicate that it is essential to investigate whether there are in animal tissues mechanisms which convert the non-morphinan reticuline by intramolecular coupling to give the morphinan salutaridine, as occurs in *P. somniferum*. A full analysis of the fact that this conversion can take place in animal tissue is reported in the paper by Goldstein's group in this issue². These authors produce convincing evidence that in rat liver reticuline can be converted to the morphinan salutaridine both *in vivo* and *in*

at the μ -receptor and of 0.31 at the κ -receptor, and [Met]enkephalyl-Arg-Phe is also less selective, having a relative affinity of 0.60 at the μ -receptor and of 0.36 at the δ -receptor. Second, the enzymatic stabilities of non-peptide and peptide opioids interacting with the μ -receptor are also very different. Most of the peptide opioids are sensitive to the degrading action of peptidases, the exception being β -endorphin; in contrast, morphine is much more stable. We still know very little about the physiological role of endogenous morphine and codeine and their correlation with the functions of opioid peptides. □

Table 1 Morphine concentration (pmol g⁻¹) in some rat tissues after intravenous injection of saline or three morphinans (from ref. 7)

Tissue	Saline	Salutaridine (31 nmol g ⁻¹)	Thebaine (10 nmol g ⁻¹)	Codeine (17 nmol g ⁻¹)
Brain	0.026	0.059	0.096	16
Liver	0.011	0.136	0.150	115
Intestine	0.017	0.099	0.112	182

Conversion to free morphine is much less with salutaridine or thebaine than with codeine.

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