New developments in opioid receptors ligands

Investigaciones actuales en ligandos de receptores opiáceos

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ABSTRACT

Relevant developments have been achieved in the last twenty years in the search for opioid agonists and antagonists with selectivity for each receptor subpopulation. Recently, new benzomorphan derivatives have been synthesized and compounds with substituted cyclopropylmethyl functionalities at N-1 position showed high affinity and selectivity for κ opioid receptor subpopulations. MPCB and CCB were selected as specific κ agonists. The affinity of CCB was two-fold the USO,488H one. Mixed peptide-heterocyclic compounds have been derived from these compounds and important informations on binding processes of κ ligands have been obtained.

Key words: Opioid Agonists and Antagonists. Selective Ligands. Bivalent Ligands. Message-Address Hypothesis.

RESUMEN

En los últimos veinte años se ha llevado a cabo una intensa investigación sobre la búsqueda de agonistas y antagonistas selectivos de cada subtipo de receptores opiáceos. Recientemente, se han sintetizado nuevos derivados benzomorfánicos y compuestos con restos ciclopropilmetílicos sobre N-1 que muestran alta afinidad y selectividad por los receptores κ. Los compuestos MPCB y CCB se han seleccionado como agonistas específicos κ. La afinidad de CCB es dos veces mayor que la del compuesto USO,488H. De estos compuestos se han derivado interesantes compuestos con estructura de peptido-heterocido y se han obtenido importantes informaciones sobre los procesos de binding a los receptores κ.

Palabras clave: Agonistas y antagonistas opiáceos. Ligandos selectivos. Ligandos bivalentes.

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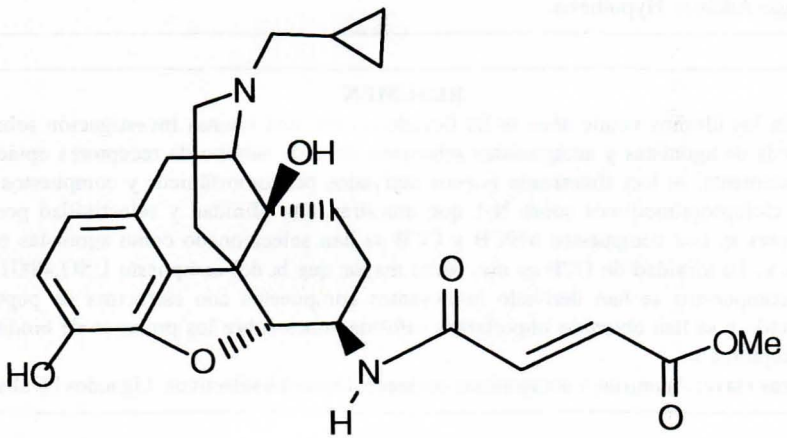
INTRODUCTION

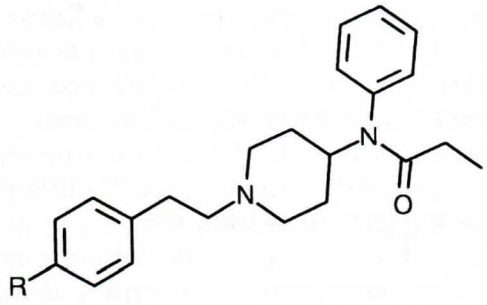
One of the key problem in Medicinal Chemistry is selectivity. Selective molecules can be used as pharmacological tools to investigate the mechanism of action and the physiological role of endogenous ligands and might possess reduced side effects when used as drugs. Nevertheless, in many cases the design of selective compounds is complicated by the existence of multiple receptor subpopulations.

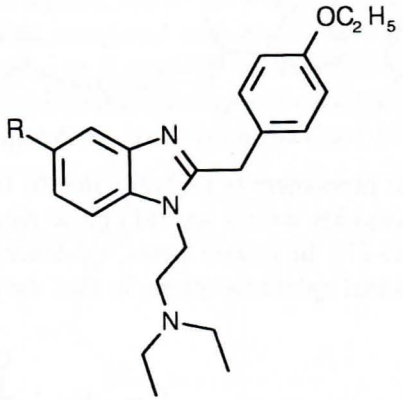
Focusing our attemption on opioid ligands, as there are at least three opioid receptors (named μ, κ and δ) (1), the search in the last two decades has been totally devoted to the design and the synthesis of selective agonists and antagonists for each type of receptor. Although several different approaches have been used by scientists to develop selective compounds, all recognize to morphine congeners an important role of peptidomimetics, as they mimic suitable relative conformations of essential functional groups present in the structure of the N-terminal fragment of endogenous ligands.

Initial studies in the field started with the elegant work of Portoghese and Takemori (2), which at the end of the seventies developped a very selective and long lasting antagonist for μ receptor subpopulation: β-FNA (3), an alkylating agent derived from β-naltrexamine capable of preventing the binding of morphine for 3-4 days to both GPI and MVD smooth muscle preparations.

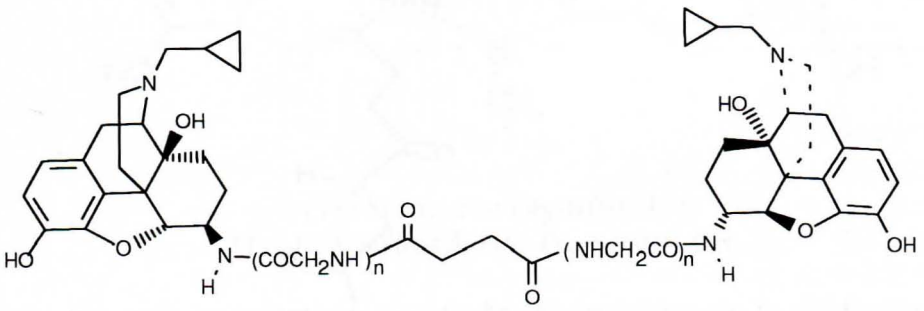


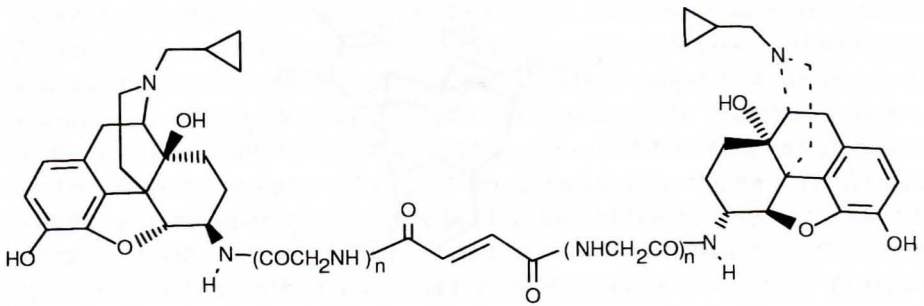
Extending this approach, Rice *et al.* synthesized several μ, κ and δ affinity labels using isothiocyanate, bromoacetamido or methylfumaramido groups as alkylating functions on fentanyl, etonitazene or endo-ethenotetrahydrooripavine skeletons (4). A different approach was later used to develop κ selective antagonists: the so called Bivalent Ligand Approach (5, 6). The synthesis and SAR studies

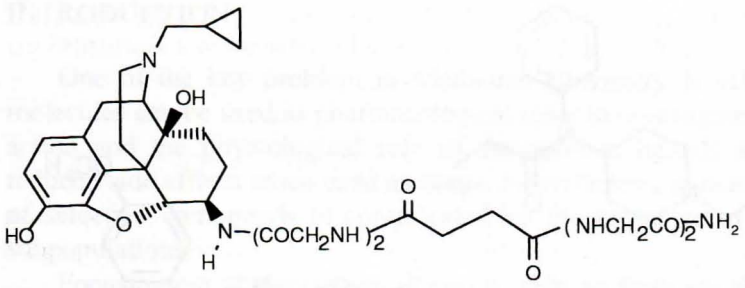




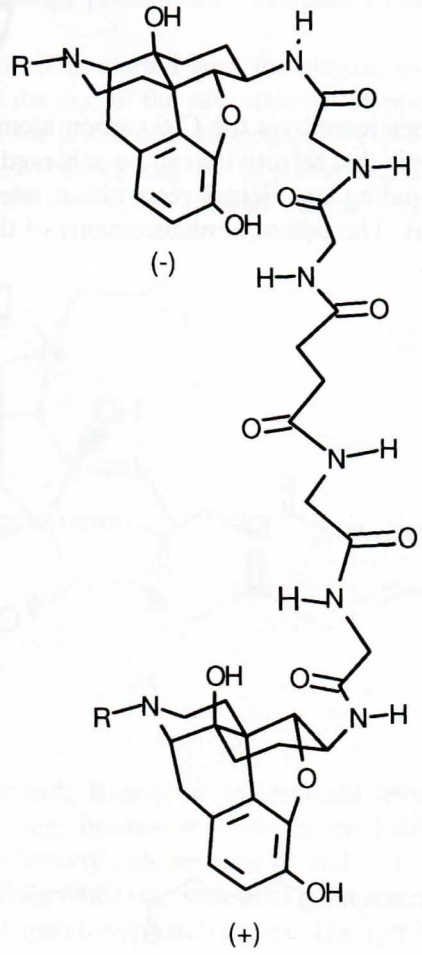
of dimeric naltrexamines joined via the C-6 carbon atom through a connecting unit led to the hypothesis that selectivity can be achieved using suitable spacers able to prevent the binding on vicinal recognition sites of particular opioid receptor subpopulations. The potency enhancements of the dimers compared to





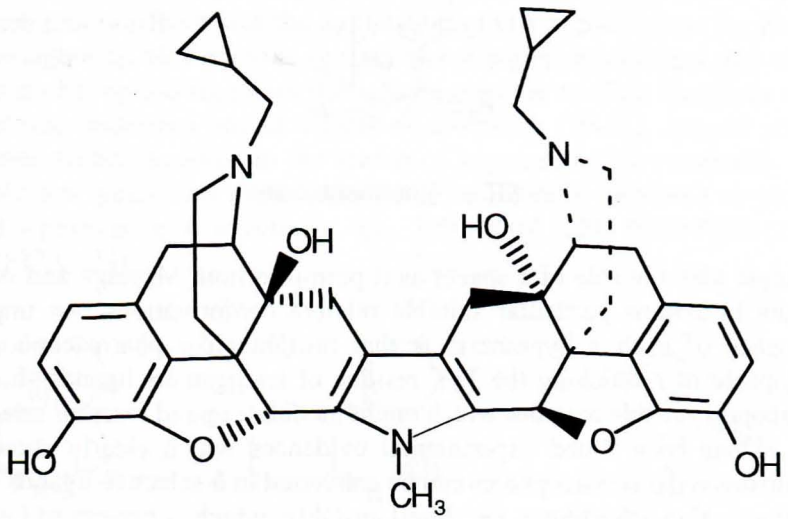


the monomers is probably due to favourable binding interactions of the second monomer with a second opioid receptor or with a proximal non-opioid binding site (7). In certain cases, evidence was achieved that these compounds bridge vicinal opioid receptors. In fact, the dimeric ligand with both (–)-pharmacophores



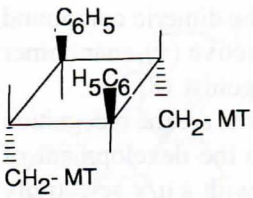
exhibited a considerable higher potency enhancement than the dimeric compound obtained joining an opioid active (–)-enantiomer with an inactive (+)-enantiomer when compared with the monovalent monomeric (–)-antagonist (8, 9).

These results, supporting the hypothesis that vicinal receptors are the recognition sites involved in the bridging of the bivalent ligand, led to the development of nor-BNI (10, 11), a very selective kappa opioid antagonist with a μ/κ selectivity ratio of 25 (naltrexone 0.2). Dimerization of μ or δ peptidic ligands has also been attempted, and informations regarding the active conformations of endogenous ligands when bind different receptor subpopulations have been obtained (see, for example, 12-15).

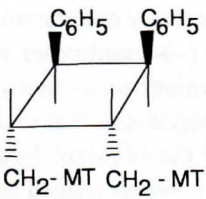


(Tyr-D-Met-Phe-Gly-NH-CH_2-)$_2$
(Tyr-D-Ala-Phe-Gly-Tyr-Pro-Ser-NH-CH_2-)$_2$

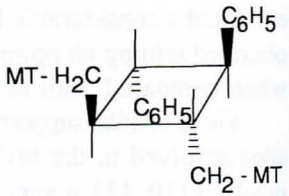
Recently, important non peptidic ligands have been obtained following the Message-Address hypothesis (16), which attempted to rationalize the ligand-receptor recognition processes. The hypothesis starts from the observation that, despite a common N-terminal tetrapeptide fragment, endogenous opioid peptides activate different receptor subpopulations. It has been suggested that the common sequence contains a "Message" capable of activating the transductional process of the receptor complex. On the contrary, the different C-terminal fragments confer different receptor selectivity by containing in suitable conformations additional functional groups specific for the interaction with certain opioid receptor subpopulations. In this model the conformation of the Phe^4 residue seems to play a key role in inducing selectivity, while the Gly^2-Gly^3 aminoacidic



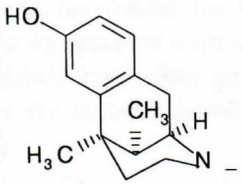
10, ε-PMTC



11, β-PMTC

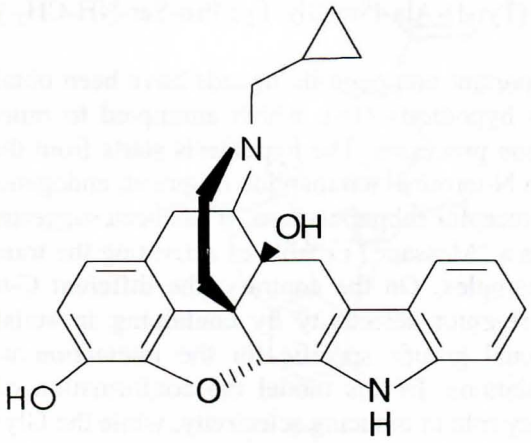


12, δ-PMTC



MT = (-)-normetazocine

chain might play the role of a spacer as it permit to both Message and Address fragments to assume particular suitable relative conformations. An important consequence of such a hypothesis is that morphinelike pharmacophores, as being capable of mimicking the Tyr[1] residue of endogenous ligands should be able to support suitable moieties which might modulate opioid receptor selectivity. In fact, it has been found experimental evidences which clearly shows that morphine derived pharmacophores can be converted in δ-selective ligands simply connecting in C-6 a δ-address, i.e., Phe-Leu-OMe, which is present in Leu-Enk, by means of a suitable spanner chain. Analogously, the same pharmacophore



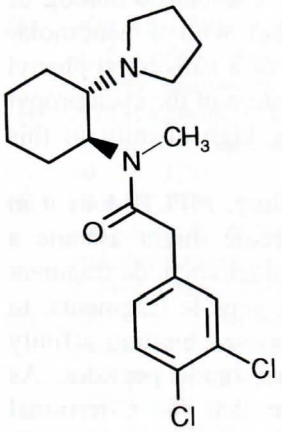
afforded selective κ-ligands by connecting in C-6 a κ-address such as Phe-Leu-Arg-Arg-Ile-OMe, which is present in Dyn-A, a κ selective endogenous ligand (17).

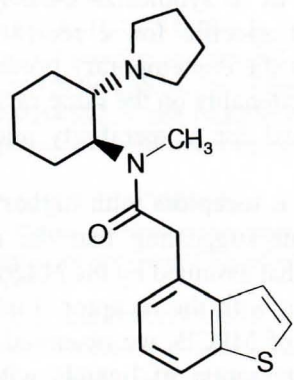
The design of selective opioid ligands based on this hypothesis has been used by several groups. Portoghese and coworkers (18) obtained δ-selective antagonists, thus confirming the key role of the phenyl ring conformation in driving the selectivity toward certain receptor subpopulations. Some naltrexone derivatives, such as NTI, resulted very potent and selective δ-antagonists.

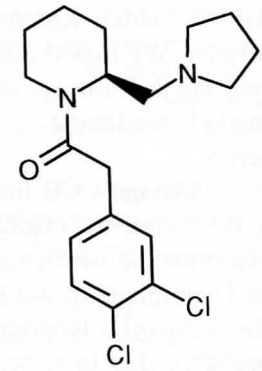
Conformational analysis in solution and in the solid state, together with experiments of molecular dynamics, evidentiated a possible conformation of a pharmacophore required for binding with delta sites. Also δ-1 and δ-2 selective molecules came from these studies.

Much more difficult was the application of this hypothesis to the design of selective agonists. However, as several recent papers established that with the activation of κ opioid receptors it might be possible to elicit analgesia without some of the undesired opioid effects of morphine (19-21), several scientists have been deeply involved in the search of kappa selective componds.

Only few years ago some compounds structurally unrelated to morphine showed significative κ selectivity, i.e., U50,488H (22), PD117302 (23) and BRL52537A (24).



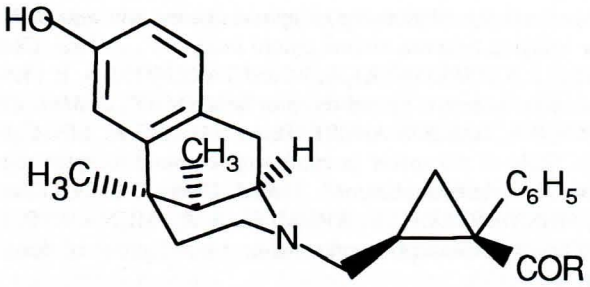




It has been suggested that a key role for selectivity was played by the relative distance and the particular relative conformation of the functional groups mimicking the functionalities present in Tyr^1 and Phe^4 residues. To better explore this hypothesis, my research group designed and synthesized a series of phenyl carboxyesters derived from N-(cyclopropylmethyl)nor-metazocine (25). The rationale for this approach was based on some simple assumptions: (i) conformational analysis of κ selective ligands suggest that the optimal distance

between the basic nitrogen and the phenolic ring is 6-8 Å; (ii) SAR analysis on U50,488H analogs put in evidence stringent relative conformational requirements for the phenyl ring which mimics the Phe[4] residue by respect to the basic nitrogen functionality; (iii) a hydrogen-bonding functionality seems to be important for binding with an accessory recognition site close to that for the aromatic ring. The choice of (–)normetazocine as a pharmacophore has been made on the assumption that several benzomorphan derivatives although possessing μ preference are also κ agonist. It was then possible that normetazocine, although a suitable pharmacophore for both μ and κ receptors may be switched to κ selectivity only adding suitable substituents supporting a phenyl ring and an H-bonding functionality with optimal distance and relative conformation by respect to the phenolic ring and the basic nitrogen. Molecular modelling studies on potential target compounds induced us to select the cyclopropylmethyl group as a spacer. The carboxyester group was introduced to probe for an accessory site on the receptor capable of interacting with an H-bonding functionality and to have informations on the stereospecificity of the ligand-receptor interaction. Among the synthesized compounds, MPCB resulted a specific κ agonist with an affinity for κ receptors 1/12 times lower than U50488H (25). It was possible to hypothesize a similar approach to κ receptors for both benzomorphan derivatives and arylacetamides and perhaps the same receptor site. On the basis of these results, the optimization process of the lead brought us to synthesize CCB (26), a 4'-chloro analog of MPCB, which resulted still specific for κ receptors but with a nanomolar affinity. We hypothesize that the contemporary presence of a subsituted phenyl ring and the carboxyester functionality on the same carbon atom of the cyclopropyl methyl substituent is essential for κ specificity and for high affinity in this series.

Although CCB binds to κ receptors with higher affinity, MPCB does it in a stereospecific manner, thus suggesting that the molecule might assume a conformation which mimics that assumed by the N-terminal tetrapeptide fragment of Dynorphins in the binding with the receptor. Linking peptide fragments, to the carboxylic functionality of MPCB, we observed improved binding affinity probably due to a better interaction of ligands with membrane peptides. As specificity is still mantained, we can thus hypothesize that the C-terminal fragment of Dynorphin is essential not for binding, but for helping the tetrapeptide fragment in assuming the right conformation for the binding with kappa receptors subpopulations (27).



R = Leu-Arg-Arg-Ile-OCH_3 (Dyn_{5-8})
Gly-Arg-Arg-Ile-OCH_3
Arg-Arg-Ile-OCH_3
OCH_3 (MPCB)

REFERENCES

(1) RAYNOR, K., KONG, H., CHEN, Y., YASUDA, K., YU, L., BELL, G. I. and REISINE, T. (1994): "Pharmacological Characterization of the cloned κ-, δ-, and μ- opioid receptors", *Molecular Pharmacol.,* **45:**330-334.

(2) PORTOGHESE, P. S., LARSON, D. L., SAYRE, L. M., FRIES, D. S. and TAKEMORI, A. E. (1980): "A novel opioid receptor site directed alkylating agent with irreversible narcotic antagonistic and reversible agonistic activities", *J. Med. Chem.,* **23:**233-234.

(3) PORTOGHESE, P. S., LIPKOWSKI, A. and TAKEMORI, A. E. (1987): "Binaltorphimine and norbinaltorphimine, potent and selective κ-opioid receptor antagonists", *Life Sci.,* **40:**1287-1292.

(4) BURKE, T. R., Jr., BAJWA, B. S., JACOBSON, A. E., RICE, K. C., STREATY, R. A. and KLEE, W. A. (1984): "Probes for Narcotic receptor mediated phenomena. 7. Synthesis and pharmacological properties of irreversible ligand specific for μ or δ opiate receptors", *J. Med. Chem.,* **27:**1570-1574.

(5) EREZ, M., TAKEMORI, A. E. and PORTOGHESE, P. S. (1982): "Narcotic antagonistic potency of bivalent ligands which contain β-naltrexamine. Evidence for bridging between proximal recognition sites", *J. Med. Chem.,* **25:**847-849.

(6) PORTOGHESE, P. S., RONSISVALLE, G., LARSON, D. L., YIM, C. B., SAYRE, L. M. and TAKEMORI, A. E. (1982): "Opioid agonist and antagonist bivalent ligands as receptor probes", *Life Sci.,* **31:**1283-1286.

(7) PORTOGHESE, P. S., LARSON, D. L., SAYRE, L. M., YIM, C. B., RONSISVALLE, G., TAM, S. W. and TAKEMORI, A. E. (1986): "Opioid agonist and antagonist bivalent ligands. The relationship between spacer lenght and selectivity at multiple opioid receptors", *J. Med. Chem.,* **29:**1855-1861.

(8) PORTOGHESE, P. S., RONSISVALLE, G., LARSON, D. L. and TAKEMORI, A. E. (1986): "Synthesis and opioid antagonist potencies of naltrexamine bivalent ligands with conformationally restricted spacers", *J. Med. Chem.,* **29:**1650-1653.

(9) PORTOGHESE, P. S., LARSON, D. L., YIM, C. B., SAYRE, L. M., RONSISVALLE, G., LIPKOWSKI, A. W., TAKEMORI, A. E., RICE, K. C. and TAM, S. W. (1985):