Protection of endogenous enkephalins by PL37, a dual NEP/APN inhibitor induces potent analgesic responses p.o. in neuropathic pain.

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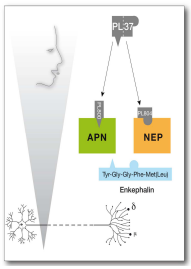
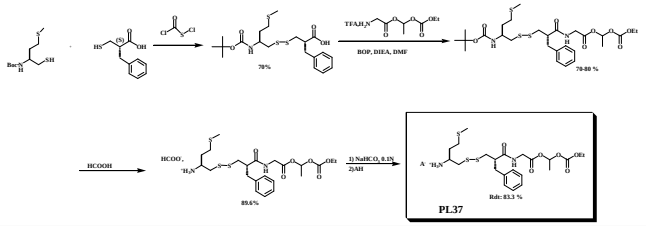
1. Introduction

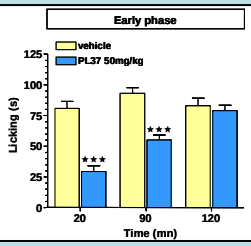
The opioid peptides Met-enkephalin and Leu-enkephalin, YGGFM(L), are inactivated by two zinc-metallopeptidases, NEP (EC 3.4.24.11) [1] and aminopeptidase N, APN (EC 3.4.11.2) [2] providing the opportunity to design enzyme inhibitors as new physiologically active analgesics acting selectively in conditions of phasic enkephalin release.
Inhibition of only one of these enzymes does not increase the endogeneous enkephalin concentration to a level sufficient to induce strong analgesic responses [3]. Therefore, we have proposed the concept of "dual inhibitors" corresponding to compounds blocking both NEP and APN [4,5]. These dual inhibitors are expected to provide new analgesics devoid of the major side effects elicited by morphine and surrogates. Moreover, the opioid systems (enkephalins, opioid receptors and inactivating enzymes) are present at the nociceptor level where they are expected to control neuropathic and neuroinflammatory pain.
Accordingly several types of dual inhibitors have been synthetized [6] and were shown very active by systemic route. The aim of this study was to design orally active compounds and this was obtained by introducing ester groups improving the water solubility and recognizing a transporter at the intestinal barrier level. Synthesis and pharmacological properties of one of these dual inhibitors, ready for phase I clinical trial, is reported here.



2. Chemistry

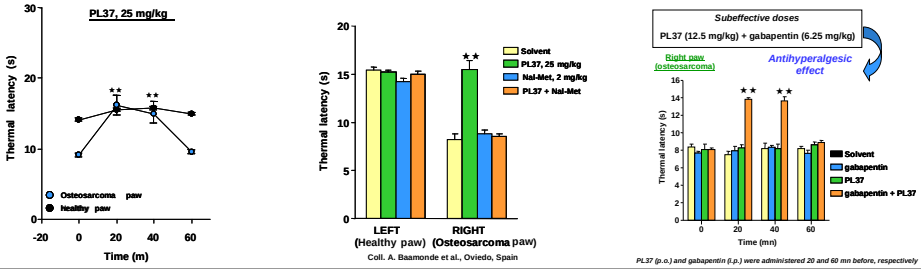


3. Formalin test



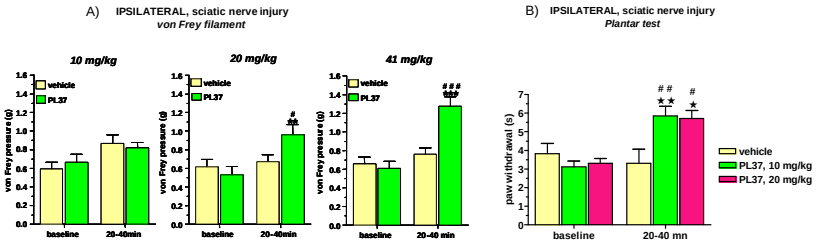
PL37 is still active 90 mn after oral administration in the early phase of the formalin test (peripheral origin of the nociceptive stimulus)

4. Bone cancer pain model



PL37 (25 mg/kg, p.o.) inhibits thermal hyperalgesia by protecting at the periphery the enkephalins acting on μ-opiate receptors since:
-naloxone-methiodide (with limited access to the CNS) and cyprodime (selective μ-opioid antagonist) block this antihyperalgesic effect.
-withdrawal latencies measured in the contralateral paw are not modified.
PL37 induces thermal antihyperalgesic effects at lower doses (12 mg/kg) in the presence of a subeffective dose of gabapentin.

5. Seltzer model



Antiallodynic (A) and antihyperalgesic (B) effects of oral PL37 in a peripheral neuropathic pain model in mice.

PL37 (20-41 mg/kg) inhibited tactile allodynia and thermal hyperalgesia induced by partial sciatic nerve ligature in mice (tests were performed 12 to 14 days after surgery). PL37 had no effect on the contralateral paw or the sham-operated paw at the higher dose tested.

6. Conclusion

Introduction of esters able to be recognized by transporters located at the intestinal barriers led to PL37 [7], an optimized dual inhibitor which is orally dose-dependent (12.5-100 mg/kg) active in the predictive animal models of neuroinflammatory and neuropathic pains (bone cancer pain, formalin, diabetic rat model, vincristine-induced neuropathy model, partial sciatic nerve ligature).
The suppression of these nociceptive stimuli is completely reversed by naloxone methiodide at a dose (2 mg/kg ip) when this antagonist is unable to enter the brain.
Finally, particularly in model of cancer-pain, the action of PL37 is synergistically potentiated by gabapentin. Preclinical studies have shown a remarkable safety profile and the absence of morphine major drawbacks (i.e. no tolerance, no constipation etc …,after repeated administration).
Therefore, a Phase I clinical study using the oral route is planned in the next months.

References
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