NEW ENDOMORPHIN-2 DERIVATIVES CONTAINING TRITIUM LABELLED 2-AMINOCYCLOPENTANECARBOXYLIC ACID AND 2-AMINOCYCLOHEXANECARBOXILIC ACID

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Endomorphin-1 (H-Tyr-Pro-Trp-Phe-NH_2) and endomorphin-2 (H-Tyr-Pro-Phe-Phe-NH_2) were revealed in the 90s as opioid ligands which can bind selectively to the μ-receptor[1]. Although their stability against proteolytic enzymes is bigger than that of the other endogenous opioid peptides for example enkephalins, the half-life of these endogenous peptides is short to produce long-lasting analgesia. To obtain a proteolyticly more stable analogues with high specificity to the μ-receptor we have synthesized endomorphin-2 analogues containing 2-aminocyclopentenecarboxylic acid (H-Tyr-ΔACPC-Phe-Phe-NH_2) and 2-aminocyclohexenecarboxylic acid (H-Tyr-ΔACHC-Phe-Phe-NH_2) based on a Boc-strategy using racemic cis-ACPC and cis-ACHC by solid phase peptide synthesis. The diastereomeric peptides were separated by RP-HPLC. The configuration of the ΔACPC and ΔACHC in the peptides was determined by RP-HPLC after the hydrogenation of all peptide isomers with comparison of earlier made standard peptides (H-Tyr-(1S,2R)ACPC-Phe-Phe-NH_2), (H-Tyr-(1R,2S)ACPC-Phe-Phe-NH_2 H-Tyr-(1S,2R)ACHC-Phe-Phe-NH_2) and (H-Tyr-(1R,2S)ACHC-Phe-Phe-NH_2,).

Previous studies have shown that the (1S,2R)ACPC and the (1S,2R)ACHC containig derivatives bind with higher affinity and specificity to the μ-receptor than the (1R,2S)ACPC and the (1R,2S)ACHC containing isomers [2]. The suitable dehydro derivatives were tritiated using tritium gas in the presence of $PdO/BaSO_4$ catalyst. We have got two saturated derivatives containing tritium in the ring of prolin mimetics. The crude tritiated peptides were purified by RP-HPLC using radioactive detector. First we have studied the binding properties of ^{3}H-(1S,2R)ACHC-endomorphin-2 derivative in rat brain membrane. The ligand showed much higher affinity to the μ-receptor than to the δ- or to the κ-receptors.

The binding studies of ^{3}H-(1S,2R)ACPC-endomorphin-2 derivative are in progress. We hope that these results will help us to better understand the structural requirements of opioid binding.

Supported by OTKA T04514 and RET 08/2004.

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