A NEW CLASS OF HIGHLY POTENT AND SELECTIVE ENDOMORPHIN-1 ANALOGUES CONTAINING α-METHYLENE-β-AMINOPROPANOIC ACIDS (MAP)

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Presented by Celeste Alverez
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J. Med. Chem. **2012**, *55*, 6224.

The opioid system

- One of the most important systems for analgesia (pain relief)
- Composed of 4 subtypes of receptors:
  - μ (MOR), κ (KOR), δ (DOR), Nociceptin (NOP)
- Ligands:
  - Endogenous peptides:
    - Dynorphins, Enkephalins, Endorphins, Endomorphins, and Nociceptin
  - Exogenous:
    - Morphine, heroin, hydrocodone, codine, fentanyl, methadone, ect.

Annu. Rev. Biochem. **2004**, *73*, 953.

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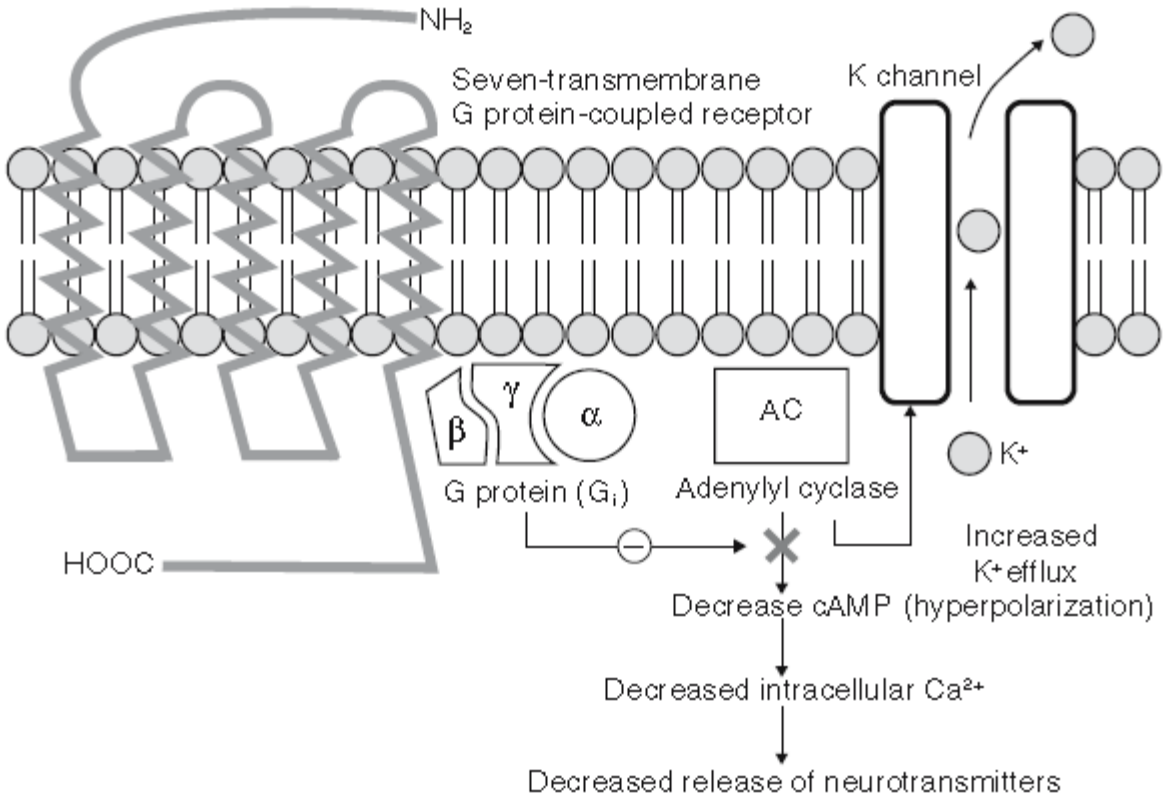
Mechanism of opioid action



Clin. Rheumatol. **2006,** *25* (Suppl 1), S9.

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Negative effects of opioids

- Can occur with both endogenous and exogenous opioids
- Side effects:
  - Drowsiness
  - Nausea
  - Muscle spasms
  - Difficulty urinating
  - Constipation
  - Addiction
  - Respiratory depression

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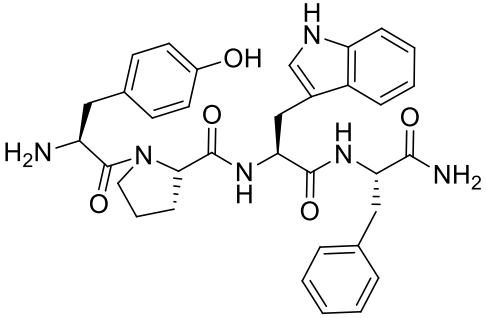
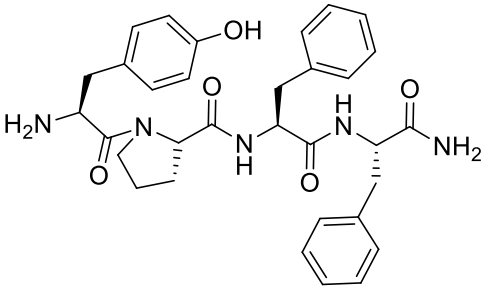
Endomorphins

- 2 types: endomorphin-1 and endomorphin-2



endomorphin-1 (EM-1)



endomorphin-2 (EM-2)

- Have analgesic properties with less undesired effects
  - Less potential for addiction
  - Less potential for respiratory depression
  - Less potential for cardiovascular complications

Med. Res. Rev. **2012**, *32*, 536.

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Endomorphins as drugs

- Obstacles to drugability of EMs:
  - Not orally avaliable
  - Short duration of action
  - Poor metabolic stability
  - Poor blood-brain barrier permability/Poor CNS avaliabilty
- Attempted solutions:
  - Unnatural amino acid substitution (D-/β-amino acids, amino acid mimetics, alkylated amino acids)
  - Cyclization
  - Glycosylation
  - Conjugation to transportable lipids

Med. Res. Rev. **2012**, *32*, 536.

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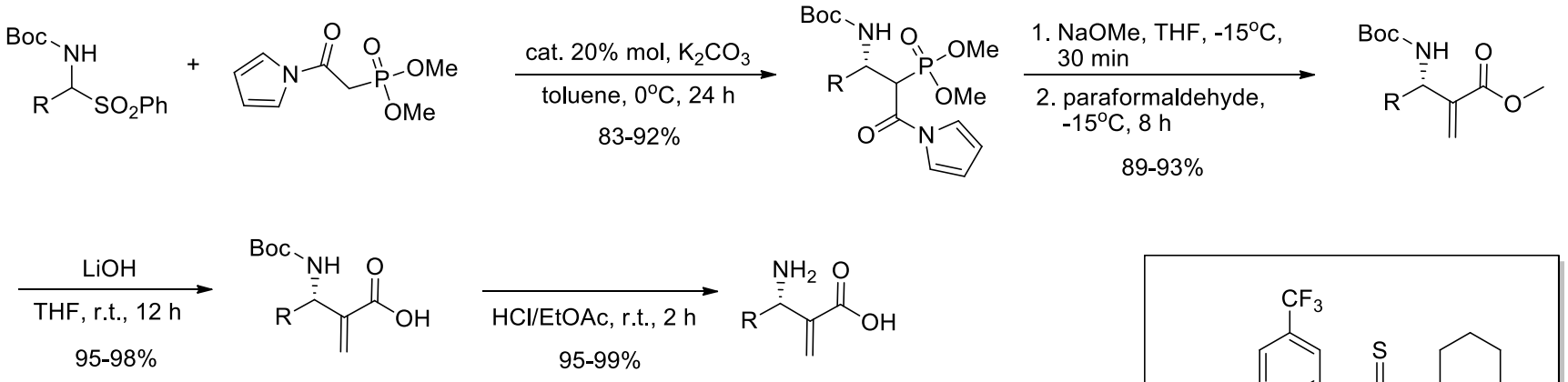
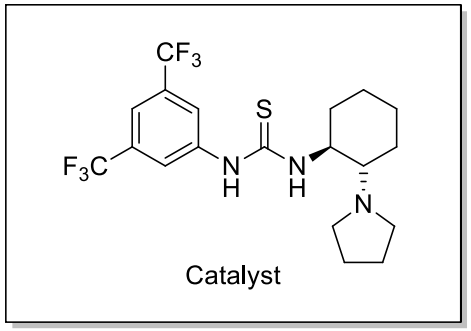
Synthesis of α-methylene-β-amino acids (Map)





	-R		-R
(Ph)Map		(Piperonyl)Map	
(4-FPh)Map		(2-Furyl)Map	
(4-ClPh)Map		(3-Furyl)Map	
(3-ClPh)Map		(1-Naphthyl)Map	
(2-ClPh)Map		(2-Naphthyl)Map	
(4-MeOPh)Map			

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Radioligand binding and selectivity

Table 2. Opioid Receptor Binding Affinities and in Vitro Pharmacological Activity of EMs and Analogues

peptide	sequence	K_i^{μ} (nM)[a,c]	K_i^{δ} (nM)[b,c]	selectivity K_i^{δ}/K_i^{μ}	IC_{50} (nM)[d] GPI	IC_{50} (nM)[d] MVD	MVD/GPI[e]
1	Tyr-Pro-Trp-Phe-NH_2	2.60 ± 0.21	6080 ± 640	2338	14.1 ± 1.7	30.4 ± 2.6	2.2
2	Tyr-Pro-Phe-Phe-NH_2	3.20 ± 0.13	6420 ± 330	2006	9.33 ± 1.12	21.6 ± 3.4	2.3
3	Tyr-Pro-(Ph)Map-Phe-NH_2	103 ± 2	59290 ± 5680	576	20.9 ± 2.37	>10000	
4	Tyr-Pro-Trp-(Ph)Map-NH_2	0.535 ± 0.076	56010 ± 5180	104692	6.81 ± 0.80	7.53 ± 1.22	1.1
5	Tyr-Pro-(Ph)Map-(Ph)Map-NH_2	15.7 ± 0.4	10980 ± 1680	699	38.1 ± 1.2	166 ± 34	4.4
6	Tyr-Pro-Trp-(4-FPh)Map-NH_2	13.7 ± 0.9	17040 ± 2050	1244	31.5 ± 1.5	130 ± 14	4.1
7	Tyr-Pro-Trp-(4-ClPh)Map-NH_2	7.12 ± 1.05	10810 ± 1340	1518	15.3 ± 3.2	36.7 ± 7.0	2.4
8	Tyr-Pro-Trp-(3-ClPh)Map-NH_2	3.49 ± 0.25	5820 ± 450	1668	7.66 ± 0.51	69.4 ± 7.4	9.1
9	Tyr-Pro-Trp-(2-ClPh)Map-NH_2	5.48 ± 0.38	14930 ± 1620	2724	16.6 ± 3.7	365 ± 14	22
10	Tyr-Pro-Trp-(4-MeOPh)Map-NH_2	4.83 ± 0.91	10200 ± 1430	2112	84.2 ± 2.0	299 ± 14	3.6
11	Tyr-Pro-Trp-(piperonyl)Map-NH_2	7.73 ± 1.02	18690 ± 1330	2418	14.3 ± 1.7	432 ± 10	30
12	Tyr-Pro-Trp-(2-furyl)Map-NH_2	0.221 ± 0.014	50010 ± 2880	226290	2.92 ± 0.31	15.8 ± 0.9	5.4
13	Tyr-Pro-Trp-(3-furyl)Map-NH_2	0.274 ± 0.066	50930 ± 6710	185876	3.94 ± 0.60	10.2 ± 1.2	2.6
14	Tyr-Pro-Trp-(1-naphthyl)Map-NH_2	26.0 ± 3.5	84680 ± 10490	3264	33.9 ± 6.2	84.4 ± 6.8	2.5
15	Tyr-Pro-Trp-(2-naphthyl)Map-NH_2	27.4 ± 0.8	84850 ± 9650	3097	18.2 ± 3.8	141 ± 6	7.7

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In vitro activity

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Inhibition of cAMP release

Table 3. Functional Activity of EMs and Analogues[a]

peptide	sequence	EC_{50} (nM)	E_{max} (%)
0	DAMGO	3.04 ± 0.32	98.14 ± 6
1	Tyr-Pro-Trp-Phe-NH_2	14.40 ± 0.62	83.13 ± 4
2	Tyr-Pro-Phe-Phe-NH_2	11.80 ± 0.23	82.75 ± 4
3	Tyr-Pro-(Ph)Map-Phe-NH_2	36.50 ± 2.45	70.78 ± 2
4	Tyr-Pro-Trp-(Ph)Map-NH_2	0.16 ± 0.09	97.94 ± 3
5	Tyr-Pro-(Ph)Map-(Ph)Map-NH_2	45.09 ± 4.01	60.26 ± 6
6	Tyr-Pro-Trp-(4-FPh)Map-NH_2	7.35 ± 1.02	81.45 ± 6
7	Tyr-Pro-Trp-(4-ClPh)Map-NH_2	12.00 ± 0.98	85.57 ± 11
8	Tyr-Pro-Trp-(3-ClPh)Map-NH_2	0.72 ± 0.08	92.53 ± 4
9	Tyr-Pro-Trp-(2-ClPh)Map-NH_2	0.84 ± 0.03	82.57 ± 4
10	Tyr-Pro-Trp-(4-MeO)Map-NH_2	10.91 ± 0.83	85.56 ± 3
11	Tyr-Pro-Trp-(Piperonyl)Map-NH_2	10.70 ± 1.09	83.90 ± 5
12	Tyr-Pro-Trp-(2-Furyl)Map-NH_2	0.0334 ± 0.0012	97.14 ± 5
13	Tyr-Pro-Trp-(3-Furyl)Map-NH_2	0.0342 ± 0.0018	98.73 ± 5
14	Tyr-Pro-Trp-(1-Naphthyl)Map-NH_2	72.30 ± 6.00	71.01 ± 4
15	Tyr-Pro-Trp-(2-Naphthyl)Map-NH_2	70.34 ± 4.67	67.46 ± 5

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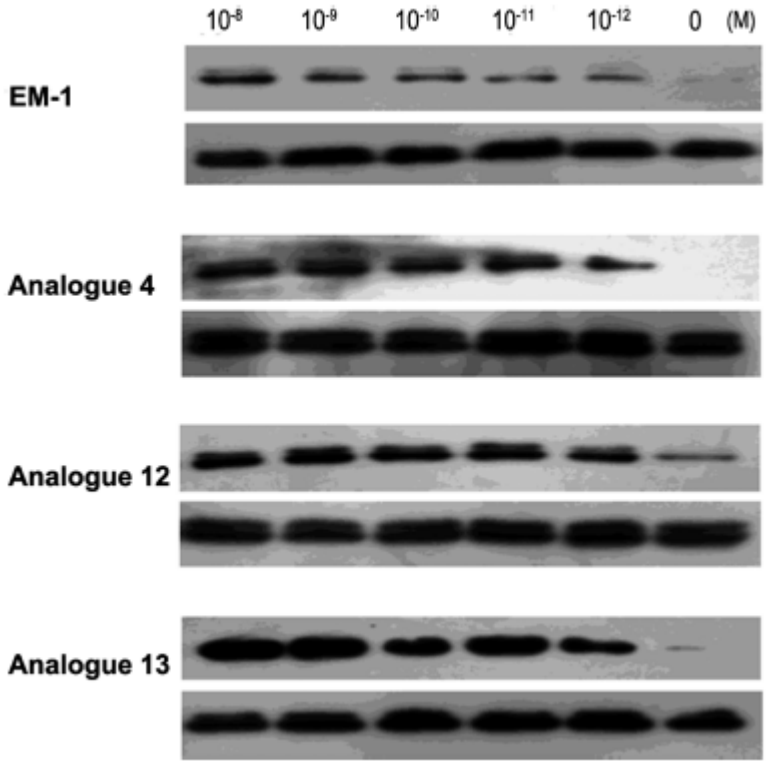
Downstream effects



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Analgesic effects

Table 4. In Vivo Antinociceptive Activities of EM-1 and Its Analogues Given icv To Produce Tail-Flick Inhibition in the Mouse

peptide	ED_{50} [a] (nmol/kg)
1	15.2 (13.1–19.3)
4	2.33 (1.74–3.03)
8	9.28 (6.66–12.5)
12	1.42 (1.11–1.88)
13	1.55 (1.09–2.06)

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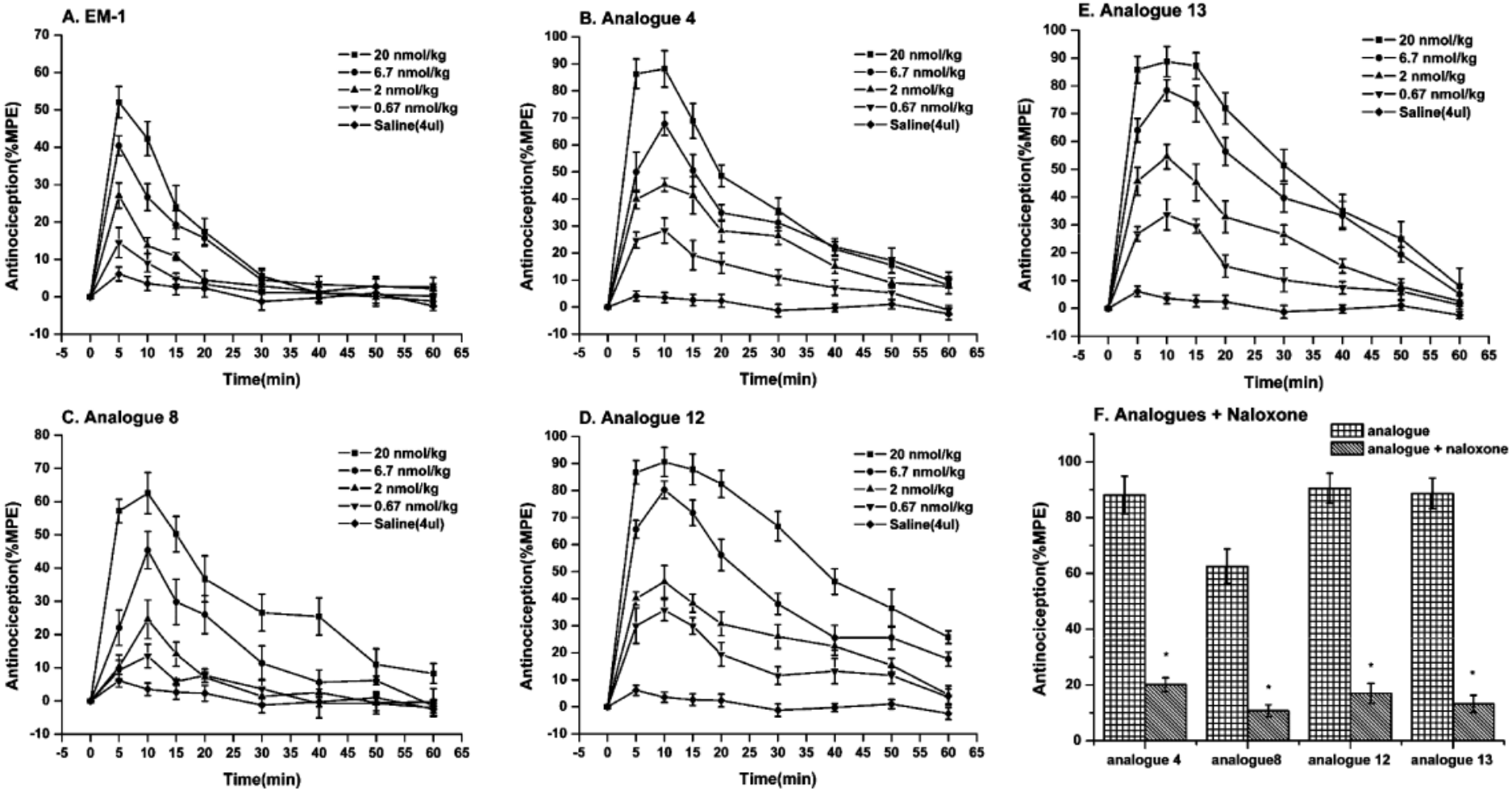
Analgesic effects



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Stability

Table 5. Half-Lives of EM-1 and Its Potent Analogues in Mouse-Brain Membrane Homogenate[a]

peptide	$100 \times k\ (min^{-1})$[b]	half-life[c]
1	4.10 ± 0.14	16.9 ± 1.2
4	1.11 ± 0.05	62.4 ± 3.1
8	0.77 ± 0.18	89.9 ± 9.3
12	0.81 ± 0.19	85.9 ± 9.2
13	0.78 ± 0.22	88.3 ± 8.2

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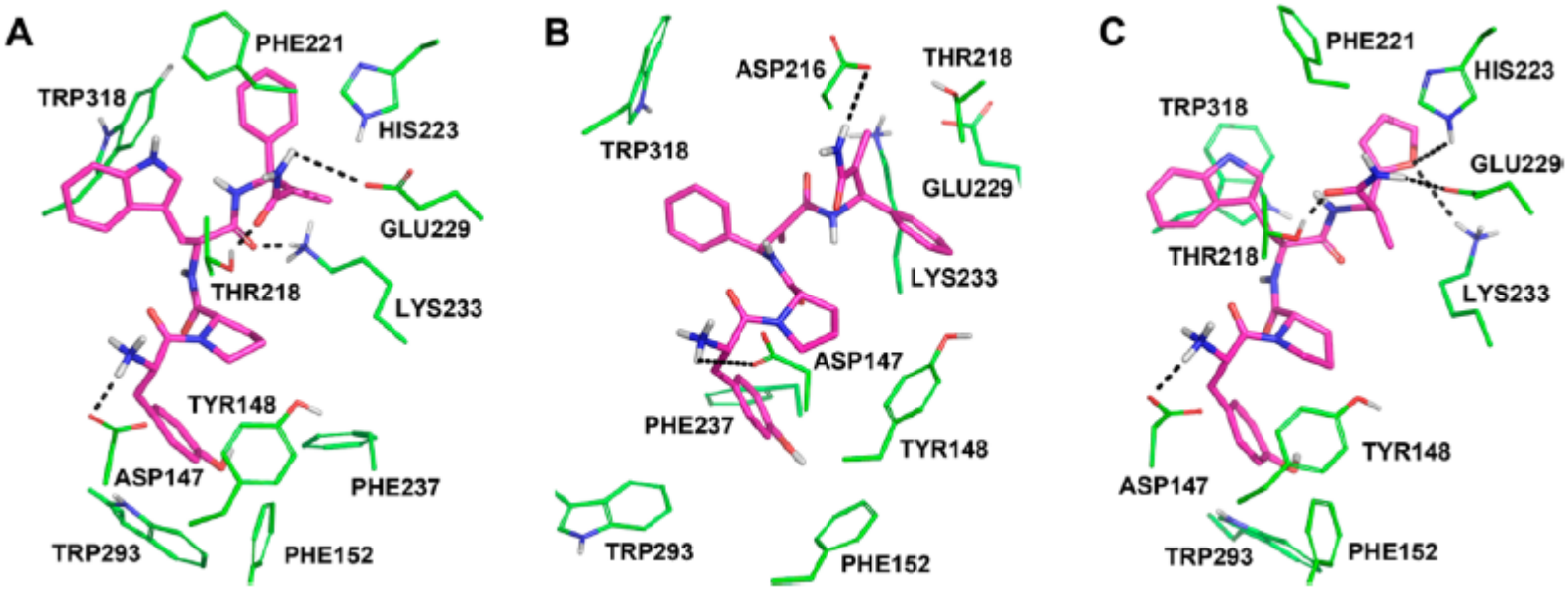
Molecular modeling



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Summary

- Developed a class of highly potent MOR selective agonists utilizing constrained unnatural β-amino acids (Map) into EM-1
- Analogues with the Map located at the 4-position (C-terminal end) were more active than those with the substitution at the 3-position
- The furan containing analogues were the most potent, effective, and stable tested

- There may be potential use of EMs modified with constrained β-amino acids as analgesics lacking some of the classical side effects of current opioid drugs

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