One Indole Ring to Rule Them All

How Modeling of Naltrindole Bound to the Delta Opioid Receptor Can Aid the Development of Novel Analgesics

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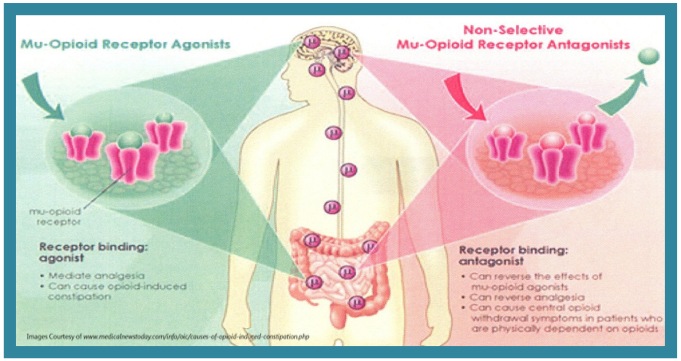
Abstract

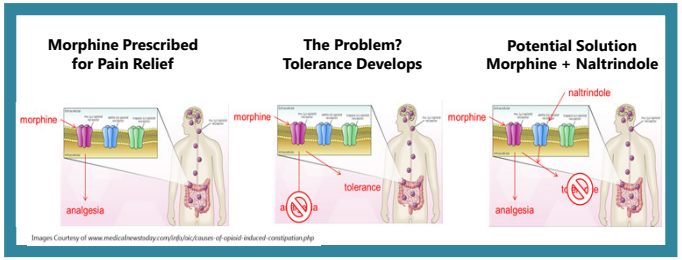
According to the Institute of Medicine, 116 million Americans currently suffer from chronic pain, costing over $500 billion annually. As such, the use of pain-killing drugs like morphine and oxycodone has increased dramatically over the past decade. Analgesic effects are produced through agonism, or activation, of the body's mu (MOR) and delta (DOR) opioid receptors, which are G-coupled protein receptors. Tolerance, the decreased analgesic effect of MOR agonists after prolonged use, is a major problem facing opioid pain management. A drug that antagonizes, or inhibits, DOR can greatly reduce the development of tolerance to MOR agonists, offering new pain therapy potentials. One example of a selective DOR antagonist is naltrindole (NTI), which has a similar structure as morphine, except for a cyclopropylmethyl group on its nitrogen substituent and a bulky indole group. The large indole ring interacts with the W318 residue on MOR but is able to bond with W284 residue on the DOR, producing DOR-selective antagonism. Co-administration of NTI with morphine represents a potential new approach to producing analgesics with less tolerance. Understanding the structure of this ligand and enzyme may lead to structure based drug design. The Marquette University High School SMART Team is modeling naltrindole bound to DOR using 3-D printing technology supported by a grant from the NIH-CTSA.

Chronic Pain[1]

- In 2011, at least 100 million adult Americans have common chronic pain conditions, a conservative estimate because it does not include acute pain or chronic pain in children.
- Pain is a significant public health problem that costs society at least $560-$635 billion annually (an amount equal to about $2,000.00 for every person living in the United States.
- In 2008 the cost to federal and state governments of medical expenditures for pain was $99 billion.

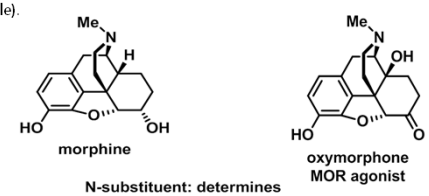
Image courtesy of www.lifebridgeblogs.org

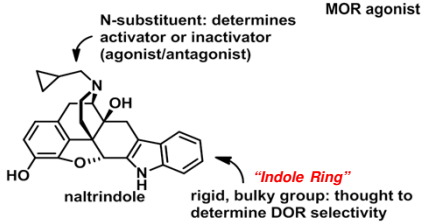




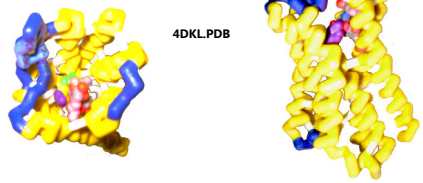
MOR and DOR

- Mu opioid receptor (μ, MOR): activation kills pain; over time, the body desensitizes to this effect, becomes tolerant.[2]
- Delta opioid receptor (δ, DOR): inhibition prevents tolerance to MOR agonists.[3]
- Naltrindole (NTI) is a selective DOR antagonist that blocks the development of tolerance to MOR agonists such as morphine.[4]
- NTI does not block any analgesic effects of morphine because it only blocks the delta receptor; it can not bind into the mu receptor binding pocket.
- The delta opioid receptor recognizes NTI and becomes inhibited. Understanding how delta recognizes antagonists is very important for when we want to design a single compound that activates mu (like morphine), and inactivates delta (like naltrindole).



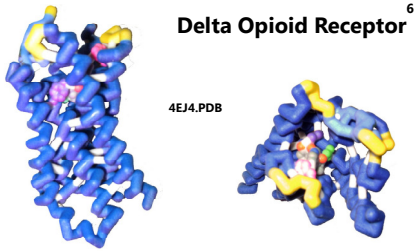


Mu Opioid Receptor[5]



Alpha Helices = Yellow
Beta Sheets = Cornflower Blue
Non-Motif Backbone = Blue
Morphinan Antagonist = CPK
Asp147 = Lime
Tyr148 = Purple
Lys233 = Hot Pink
His297 = Chocolate
H Bonds = Dodger Blue
Struts = White

Delta Opioid Receptor[6]

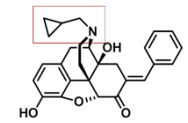


Alpha Helices = Blue
Beta Sheets = Cornflower Blue
Non-Motif Backbone = Yellow
Naltrindole = CPK
Asp128 = Lime
Tyr129 = Purple
Trp284 = Magenta
Struts = White
H Bonds = Dodger Blue
Struts = White

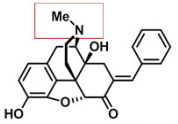
The Next Frontier: BOM (UMB246)

A single compound that simultaneously activates MOR and deactivates DOR to treat chronic severe pain could lower health care costs and benefit patients. This was the rationale behind the development of UMB246.

In 1992, the Portoghese group at the University of Minnesota made benzylidenenaltrexone (BNTX)[7], which was a DOR_1 antagonist with modest selectivity over MOR. Because most *N*-methyl opioids are mu agonists, researchers at the University of Maryland hypothesized that replacement of the *N*-group of BNTX with an *N*-methyl would produce a MOR agonist. Additionally, the benzylidene group was hypothesized to be flexible enough to allow binding to MOR. The result is benzylideneoxymorphone (BOM, UMB246), a new small-molecule MOR partial agonist/DOR antagonist structural scaffold.



benzylidinenaltrexone (BNTX)
MOR/DOR antagonist



benzylidineoxymorphone (BOM, UMB246)
MOR partial agonist/DOR antagonist

Laboratory Data[8]

Cells expressing each opioid receptor subtype were obtained from Dr. Larry Toll (SRI International, Manlo Park, CA). Competitive binding experiments were performed in the laboratory of Prof. Rae R. Matsumoto (West Virginia University). Table 1 shows UMB246 binds similarly well to MOR and DOR, and is approximately 60-fold selective over KOR.

Table 1: Percent inhibition (specific binding) of MOR, DOR, KOR.

Compound Name (UMB)	μ: K_i (nM)	δ: K_i (nM)	κ: K_i (nM)
246	17.5 ± 1.10	14.4 ± 0.65	1067 ± 39

Conclusion

Both the mu opioid receptor (MOR) and the delta opioid receptor (DOR) play a crucial role in analgesia. DOR can modulate the tolerance effect resulting from prolonged administration of opiates like morphine and oxycodone (OxyContin). In order to maintain analgesic effects, caretakers must increase the dosage of an opiate administered to a patient. This is extremely expensive and can lead to various adverse side effects such as constipation and increased risk of overdose. In order to combat these side effects, chemists and pharmacologists are attempting to develop drugs that have a lessened tolerance effect. By antagonizing, or shutting off, the DOR receptor, the tolerance effect is eliminated. Naltrindole (NTI) fills this role, but unfortunately it does not agonize, or turn on, the MOR receptor, and thus has absolutely no analgesic effect by itself. Therefore, scientists are trying to develop MOR agonist/DOR antagonist drugs that would produce pain-killing effect without tolerance. Benzylideneoxymorphone (BOM, UMB246) may serve as an example of this type of compound. The purpose of this scientific exploration is to develop a solution to the chronic pain that plagues 110 million Americans and costs our nation $500 billion annually.

References

1 Institute of Medicine Report from the Committee on Advancing Pain Research, Care, and Education. Relieving Pain in America, a Blueprint for Transforming Prevention, Care, Education and Research. The National Academies Press, 2011, pp. 17, 260.
2 Keiffer, B.L; Evans, C.J. Opioid tolerance – in search of the holy grail. Cell 2002, 108, 587-590.
3 Abdelhamid, E.E.; Sultana, M.; Portoghese, P.S.; Takemori, A.E. Selective blockage of the delta opioid receptors prevents the development of morphine tolerance and dependence in mice. J. Pharmacol. Exp. Ther. 1991, 258, 299-303.
4 Hepburn, M.J.; Little, P.J.; Gringas, J.; Kuhn, C.M. Differential effects of naltrindole on morphine-induced tolerance and physical dependence in rats. J. Pharmacol. Exp. Ther. 1997, 281, 1350-1356.
5 Manglik, A.; Kruse, A.C.; Kobilka, T.S.; Thian, F.S.; Mathiesen, J.M.; Sunahara, R.K.; Pardo, L.; Weis, W.I.; Kobilka, B.K.; Granier, S. Crystal structure of the mu-opioid receptor bound to a morphinan antagonist. Nature2012, 485, 321-326.
6 Granier, S.; Manglik, A.; Kruse, A.C.; Kobilka, T.S.; Thian, F.S.; Weis, W.I.; Kobilka, B.K. Structure of the delta-opioid receptor bound to naltrindole. Nature 2012, 485, 400-404.
7 Portoghese, P.S.; Sultana, M.; Nagase, H.; Takemori, A.E. A highly selective δ1-opioid receptor antagonist: 7-benzylidenenaltrexone. Eur. J. Pharmacol. 1992, 218, 195-196.
8 Cunningham, C.W.; Healy, J.R.; Kudrimoti, S.; Matsumoto, R.R.; Coop, A. Unpublished results (Manuscript in preparation).

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