Role of Phosphorylation in Acute Desensitization and Tolerance of the μ-Opioid Receptor
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**Background**

- There is increasing evidence that μ-opioid receptor (MOR) acute desensitization, or the rapid loss of MOR-effector coupling during sustained agonist exposure, is a critical step leading to long-term tolerance to opioids.
- Agonist-induced activation of MOR leads to phosphorylation of the intracellular region by G protein-coupled receptor kinases (GRKs) and phosphorylation of the C-terminal tail of MOR is a necessary step in acute desensitization.
- Acute desensitization is nearly abolished and cellular tolerance induced by chronic morphine treatment is reduced for phosphorylation-deficient mutant MORs in the LC of the rat.
- In this study, we will investigate what kinases are involved in MOR acute desensitization as well as whether or not more desensitization develops following longer applications of opioids.

**Methods**

- Used whole-cell voltage-clamp recordings from Locus Coeruleus (LC) neurons in acute brain slices from Sprague-Dawley rats
- LC neurons express μ-opioid receptors (MORs) that are coupled to G-coupled inwardly rectifying potassium channels (GIRKs)
- Degree of acute desensitization was determined using an EC_{50} concentration of MOR agonist following treatment with a saturating concentration of agonist for different periods
- There are similar amounts of desensitization when phosphorylation is blocked with a GRK2/3 inhibitor and when it is blocked with the total phosphorylation-deficient mutant MORs

**Acute Desensitization of the μ-opioid receptor**

Control

- ME (0.3)
- ME (2 min)
- 100 pA

Total Phosphorylation-Deficient Mutant MORs

- ME (0.3)
- ME (2 min)
- 200 pA

Compound 101 (GRK2/3 Inhibitor)

- ME (0.3)
- ME (2 min)
- 100 pA

**Desensitization after Long Incubation**

1 hr Incubation in DAMGO

- ME (0.3)
- ME (30 min)
- 200 pA

**Future Directions**

- Treat animals for 2 weeks with morphine and see if there is more cellular tolerance
- Investigate the role of PKC using PKC inhibitors

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