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Characterization of an exon 7-associated seven transmembrane C-terminal variant, mMOR-1O, of the mu opioid receptor gene (*Oprm1*) in a conditional gene targeting mouse model

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Background: A single-copy mu opioid receptor (*OPRM1*) gene generates an array of mu opioid receptor variants or isoforms through extensive alternative splicing. One set of these variants are 7 transmembrane (TM) C-terminal splice variants. Our studies using an exon 7 (E7) C-terminal truncation mouse model demonstrated the functional importance of exon 7-encoded sequences in morphine actions. In the current study, we generated a new mouse model (mE1-KO/mM1O-cKI) in which all 7TM variants are knocked out and only mMOR-1O, an E7-associated 7TM variant, is conditionally expressed via Cre-LoxP recombination under the control of endogenous exon 1 promoter.
Rationale/Significance: Investigating the roles of individual 7TM full-length C-terminal variants in mu opioid actions will advance our understanding of complex mu opioid actions and provide potential therapeutic targets for pain management.
Hypothesis: Each individual Oprm1 7TM C-terminal variant play unique roles in mediating mu opioid actions.
Result: We generated mE1-KO/mM1O-cKI mouse in C57BL/6J mice using a novel *Easi*-CRISPR approach. [^{3}H]DAMGO binding and morphine analgesia were completely lost in this model, but regained in mouse (mE1-KO/mM1O-cKI^{Cre}) after crossing with a CAG-Cre mouse. In mE1-KO/mM1O-cKI^{Cre} mice, morphine tolerance was developed much faster than in WT control mice.
Discussion: Our results indicate effectiveness of *Easi*-CRISPR in generating gene targeting mouse models, particularly with a large insertion. mMOR-1O facilitates morphine tolerance, which is consistent with the results from our E7 truncation mouse model. Further pharmacological and biochemical characterization of this mouse will provide useful information regarding the functions and mechanisms of mMOR-1O in mu opioid pharmacology.
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