

The role of alternatively spliced mu opioid receptor C-terminal tails in morphine tolerance, physical dependence and reward

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The mu opioid receptor (*OPRM1*) gene undergoes extensive alternative pre-mRNA splicing, creating an array of splice variants that are conserved from rodent to human. Of these splice variants, the carboxyl (C-) terminal 7 transmembrane (TM) variants share an identical receptor structure, but have a different intracellular C-terminal tail. We explore the role of the *OPRM1* C-terminal splice variants in morphine actions using several gene targeting mouse models truncating either all C-terminal tails or only C-terminal tails encoded by exon 4 or exon 7 in two different inbred strains, C57BL/6J (B6) and 129/SvEv. Characterizing these mice reveals divergent roles of individual C-terminal tails in various morphine actions, such as tolerance, physical dependence, locomotor activity and reward, highlighting the importance of individual intracellular C-terminal tails in mediating complex morphine actions. Truncating exon 7 (E7)-associated C-terminal tails in B6 mice (mE7M-B6) attenuated morphine tolerance and reward with no change in morphine dependence, whereas truncating exon 4 (E4)-associated C-terminal tails in B6 mice (mE4M-B6) facilitated morphine tolerance and reduced morphine dependence without affecting morphine reward. The similarity in several morphine-induced behaviors and receptor desensitization between mE7M-B6 homozygous and β -arresin2 KO mice suggest a physical and functional association of E7-associated C-terminal tails with β -arrestin2, a hypothesis further supported by our in vitro data that several mu agonists displayed greater β -arrestin bias against E7-associated variants than on the E4-associated mMOR-1. C-terminal splice variants are not limited to the *OPRM1* gene. Genomic database analysis suggests that as many as 12% of the human GPCR genes (excluding olfactory receptor genes) generate alternatively spliced intracellular C-terminal tail variants. However, the expression and function of the majority of these C-terminal variants remain unknown. Our study raises the more general question of whether 3' alternative splicing may expand the pharmacological repertoire of GPCRs in general.

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