Identification of new functional selective oxytocin-derived agonists that discriminate between individual G protein family subtypes.

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The Oxytocin receptor (OTR), like many other G-Protein Coupled Receptors (GPCRs), can couple to more than one class of G-proteins and we demonstrated that oxytocin (OT) promotes the receptor-mediated engagement and activation of $G_q$ and all the $G_{i/o}$-family members. We used a BRET biosensor to screen for functional selective ligands of the human OTR; these analogs, by targeting only one specific signal transduction pathway at a time, may be of great pharmacological and clinical relevance. Among the OT-analogues tested, a number behaved as biased agonists at different G protein subtypes. Particularly, with only one exception, all the peptides that activated $G_q$ also activated $G_{i2}$ and $G_{i3}$, but not $G_{i1}$, and none of them activated $G_{oA}$ or $G_{oB}$. Two peptides (DNalOVT and atosiban) activated only $G_{i1}$ or $G_{i3}$, but failed to recruit beta-arrestins and to cause receptor internalization. The development of a fluorescent derivative of atosiban allowed us to visualize, by in vivo confocal experiments, its in vivo binding to receptors, and to confirm the impaired ligand-induced receptor internalization. Finally, DNalOVT and atosiban inhibited cell proliferation, showing that a single $G_i$ subtype-mediated pathway is sufficient to prompt this physiological response. These analogs represent unique tools for examining the contribution of $G_{i/o}$ members in complex biological responses and open the way to the development of drugs with peculiar selectivity profiles characterized by long-lasting activity.