No. 2121

Brain Ca^{2+} Activity Involved in Gain/Loss of Happiness

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Summary

Oxytocin (OT) is a neuropeptide that is also known as the "happy hormone" or "love hormone." It regulates diverse brain function, from social cognition to emotion to appetite, and is a potential therapeutic agent for many mental illnesses. OT acts on its receptor named OT receptor, a Gq-coupled G protein coupled receptor (GPCR) that triggers intracellular Ca^{2+} signals upon stimulation. Therefore, OT-induced Ca^{2+} signaling is a key to understand how happy feelings are created in the brain. Because Ca^{2+} signaling is versatile, in other words that can be induced by a variety of stimulation molecules in our body; therefore, recording of OT specific-Ca^{2+} signals is necessary to advance our knowledge on Ca^{2+} signaling involved in happy feelings. To achieve that, development of a new technology to measure brain OT level in living animal is critical. Thus, we addressed to engineer a new OT sensor for *in vivo* real-time recording at first.

In this study, we developed an ultrasensitive fluorescent OT sensor, designated as $MTRIA_{OT}$, which was a chimera protein composed of medaka OT receptor, and an engineered green fluorescent protein (GFP)-based module. We obtained $MTRIA_{OT}$ through the three-step screening of mutant sensors in cultured cells; where amino acid residues in three important regions of the chimera protein were sequentially optimized. $MTRIA_{OT}$ shows robust fluorescence responses upon agonist binding, likely because of optimal coupling between the conformational change of an intracellular loop of the OT receptor and the environmental change of the GFP chromophore.

Having validated the basic properties of $MTRIA_{OT}$ in cultured cell, we performed *in vivo* recordings brain OT dynamics in living mice by using fiber photometry-mediated fluorescence measurements. We demonstrated that $MTRIA_{OT}$-mediated *in vivo* fluorescence recording can report artificially-evoked OT responses as well as endogenously-controlled OT signals with a fast temporal resolution. Importantly, our analysis revealed that the temporal profiles of OT signals are highly variable depending on the behavioral context of the animal, and that the dynamics can be altered by perturbations, such as administration of anesthetic drugs, food deprivation, and aging. Overall, our new tool, $MTRIA_{OT}$, opened the door to detect OT dynamics in the living brain in real-time.