

Fluorescent Sensor Motif for Designing Sensors for G-Protein Coupled Receptor Activation, PPI, and Peptide Bond Cleavage

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OVERVIEW

A novel, genetically encoded method to detect GPCR signaling, PPI, and protease cleavage events.

- Easily introduced into cell lines to produce high-throughput screening assays
- Can be tuned to target specific components of cells for enhanced data collection

BACKGROUND

G-Protein coupled receptors (GPCRs), protein-protein interactions (PPI), and protease cleavage events are critical processes that help regulate human health. These cellular events can also play a role in the development of disease. In general, GPCRs are responsible for detecting extracellular chemicals and converting that detection signal into an intracellular response. G-protein activation results in the production of various secondary messengers, which ultimately help to regulate bodily functions. Cell signaling and protein-protein interactions (PPIs) are therefore essential in living systems. Due to their importance, many tools have been developed to detect cell signaling events. Still, a need exists to improve genetically encoded methods to detect G-protein coupled receptor (GPCR) signaling and protease cleavage events.

INNOVATION

Researchers at the University of Michigan have discovered a new sensor design motif that can be developed into fluorescent sensors for the detection of signaling molecules (agonists) for G-protein coupled receptor (GPCR), protein-protein interactions (PPI), peptide-bond cleavage events, and a variety of other signaling events. This technology incorporates a circular-permuted fluorescent protein (cpFP) linked to a secondary protein that inhibits the cpFP's fluorophore maturation. When the secondary protein is removed from the cpFP as a result of a conformational change or peptide bond cleavage event, the fluorophore matures and fluoresces. The genetically encoded sensor motif can be easily introduced into cells, allowing for the creation of screening cell lines and high-throughput screening assays for drug development. This technology can also be tuned to target certain compartments of a cell, resulting in higher quality data collection. Collected data shows promising agonist detection capabilities for the mu-opioid receptor, kappa-opioid receptor, and beta-2 adrenergic receptor. Overall, this novel technology has a wide variety of potential in vitro and in vivo applications.

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