CREATIVE BIOMART®
Assay Kit

TR-FRET PPAR gamma Coactivator Assay Kit, goat

Product Information

Cat

Kit-0891

Common Name

TR-FRET

Cat.No. Kit-0891

Description

The TR-FRET PPAR gamma Coactivator Assay Kit provides a sensitive and robust method for high-throughput screening of potential PPAR gamma ligands as agonists or antagonists of ligand-dependent coactivator recruitment. The kit uses a terbium (Tb)-labeled anti-GST antibody, a fluorescein-labeled coactivator peptide, and a Peroxisome Proliferator Activated Receptor (PPAR) gamma ligand-binding domain (PPAR gamma-LBD) that is tagged with glutathione-S-transferase (GST), in a homogenous mix-and-read assay format.

Agonist mode

To run the TR-FRET Peroxisome Proliferator Activated gamma Receptor Coactivator Assay in agonist mode (to identify agonist compounds), PPAR gamma-LBD is added to ligand test compounds followed by addition of a mixture of the fluorescein-coactivator peptide and Tb-anti-GST antibody. After an incubation period at room temperature, the TR-FRET 520: 495 emission ratio is calculated and used to determine the EC50 from a dose response curve of the compound. Based on the biology of the PPAR gamma-coactivator peptide interaction, this ligand EC50 is a composite value representing the amount of ligand required to bind to receptor, effect a conformational change, and recruit coactivator peptide.

Antagonist mode

When the TR-FRET Peroxisome Proliferator Activated gamma Receptor Coactivator Assay is run in antagonist mode (to identify antagonist compounds), PPAR gamma-LBD is added to ligand test compounds followed by addition of a mixture of agonist, fluores-cein-coactivator peptide, and Tb-anti-GST antibody. The concentration of agonist used in this mode is the EC80 concentration as determined by first running the assay in agonist mode.

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Applications

Co-Factor Interaction Assay

Storage

Store in ultra-cold freezer (-68 to -85°C).

Shipping

Dry Ice

Size

800 x 20 µL assays

Kit Components

The TR-FRET PPARγ Coactivator Assay Kit contains PPAR gamma-LBD (GST) protein, fluorescently labeled TRAP220/ DRIP-2 coactivator peptide, Tb-anti-GST antibody, and buffers. Store components as indicated in the assay protocol (-80°C, -20°C, or +4°C).

Materials Required but Not Supplied

Microplate Reader

Detection method Fluorescent

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