Gastric Inhibitory Polypeptide Receptor (GIPR) ACTOne™ Stable Cell Line

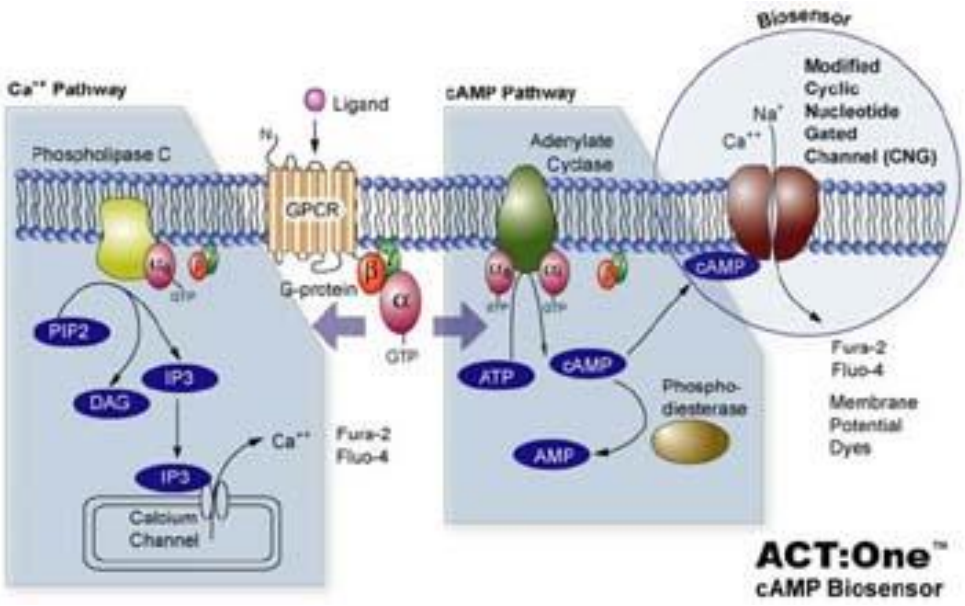
CATALOG NUMBER: CL-01-GIPR

Introduction

GIPR is G-protein coupled receptor for gastric inhibitory polypeptide (GIP), which was originally identified as an activity in gut extracts that inhibited gastric acid secretion and gastrin release, but subsequently was demonstrated to stimulate insulin release in the presence of elevated glucose. Mice lacking this gene exhibit higher blood glucose levels with impaired initial insulin response after oral glucose load. Defect in this gene thus may contribute to the pathogenesis of diabetes**.**

Description

Human GIPR ACTOne™ is a HEK-293 CNG cell line that expresses recombinant human GIPR. HEK-293 CNG cells express a modified CNG (Cyclic Nucleotide Gated) channel that opens in response to elevated intracellular cAMP levels and consequently result in ion flux (often detectable by calcium-responsive dye, Cat.# CA-C155) and cell membrane depolarization which can be easily measured with fluorescent Membrane Potential Dye (Cat.# CA-M165). The assay allows both end-point and kinetic measurement of intracellular cAMP changes with a FLIPR, or a fluorescence microplate reader.



Parental Cells

HEK-293 CNG cells (originally developed by BD Biosciences by introducing CNG in HEK-293 cells) (Cat# CL-03-PC20)

Gene/Enzyme Introduced

GIPR (Genbank Accession No. NP_000155)

Applications

- cAMP dependent human GIPR cell based assay
- cell based high-throughput screening of human GIPR inhibitors

Functional Test

- this cell line has been tested positive for GIPR specific response

- surviving rate: More than 2.5 million/vial on the second day after thawing
- the receptor specific activity is stable for 10 weeks continuous passage

Mycoplasma Contamination Test

This lot of cells has been tested and found to be free of mycoplasma contamination.

Content

- Stable cells: 1 mL (1 x 10^6 cells/mL in 70% DMEM, 20% FBS, 10% DMSO)

Growth Properties

Adherent

Cell Culture Medium

- Growth medium: DMEM-10% FBS supplemented with 250 µg/ml G418, 1 µg/ml Puromycin
- Freezing medium: 10% DMSO, 90% complete cell culture medium

Storage

Remove the frozen cells from the dry ice packaging and immediately place the cells at a temperature below -130°C, preferably in liquid nitrogen vapor, until ready for use.

Data Analysis

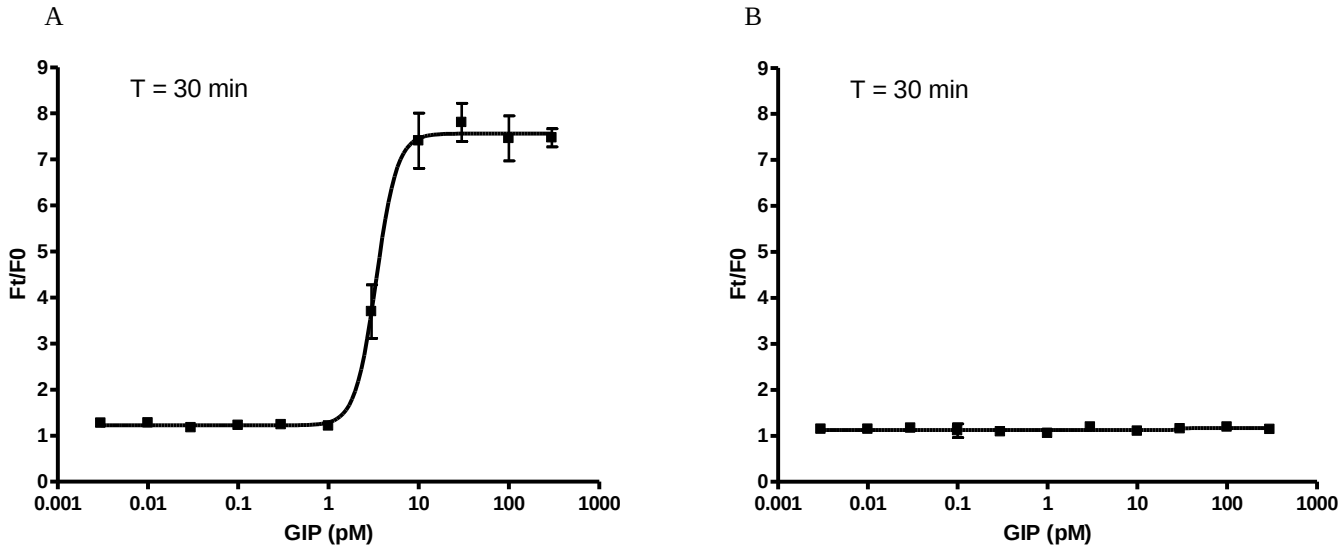


Figure 1. Response of ACT*One*™ GIPR cell line & parental cell line to GIP.

ACTOne™ GIPR cells and parental cells (Cat# CL-03-PC20) were plated overnight in 20 µl culture medium on a 384 well Biocoat plate. The next day, cells were dye-loaded with 20 µl/well of 1x Dye-loading solution (membrane potential dye kit, Cat# CA-M165). After 2 hours of incubation at room temperature, two readings were obtained prior to and 30 min after the addition of GIP. Ratios of the two readings (F/F0) are plotted in the figure.

A. Dose response curve of GIP in ACT*One*™ GIPR cell line. EC50 = 3.4 pM in the presence of PDE inhibitor Ro 20-1724, and EC50 = 12.5 pM in the absence of Ro20-1724 (data shown).

B. Parental cells do not respond to GIP.

Notice to Purchaser

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