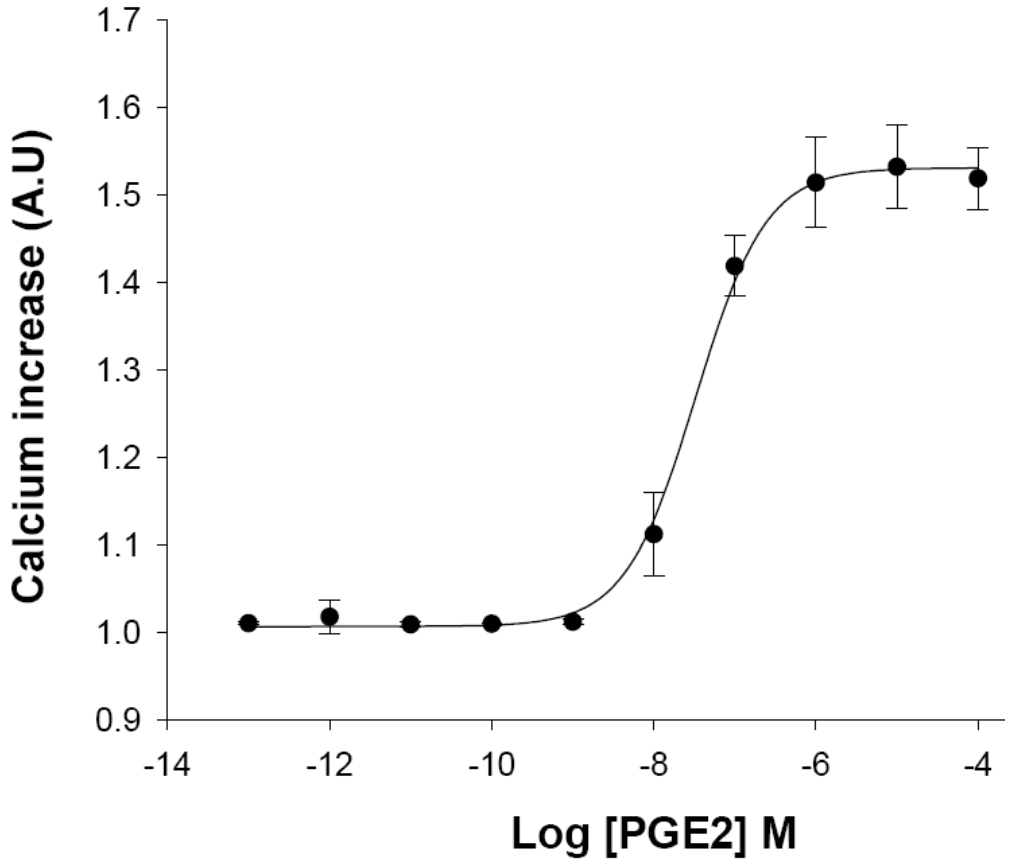
REF: P30408

HiTSeeker CELL LINES (LABEL-FREE GPCRS)

- PROSTAGLANDIN F RECEPTOR (PTGFR) CELL LINE -



Product name: FP (PTGFR) /U2OS cell line

Ec_{50} PGE2: 3.29 x 10^{-8} M

Z´: 0.78+/- 0.02

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Innoprot
Speeding up drug discovery

REF: P30408

- PROSTAGLANDIN F RECEPTOR (FP) CELL LINE -

Product Name:	PTGFR/U2OS
Official Full Name:	Prostaglandin F receptor
DNA Accession Number:	GenBank: AY337000
Host Cell:	U2OS
Format:	2 cryopreserved vials
Resistance:	G418
Size:	> 3 x 10^6 cells / vial
Storage:	Liquid Nitrogen

Assay Briefly description

Each vial of HiTSeeker PTGFR contains U2OS cells stably expressing human Prostaglandin F receptor (PTGFR) with no tag.

Innoprot's HiTSeeker PTGFR cell line has been designed to assay compounds or analyze their capability to modulate Prostaglandin F receptor. When an agonist binds to PTGFR a G protein is activated, which in turn, triggers a cellular response mediated by second messengers (Calcium).

This cell line has been validated measuring calcium increase in the cytosol. The high reproducibility of this assay allows monitoring FP activation process in High Throughput Screening.

About PTGFR

Prostaglandins are a group of lipid compounds that participate in a wide range of body functions.

The protein encoded by this gene is a member of the G-protein coupled receptor family. This protein is a receptor for prostaglandin F2-alpha (PGF2-alpha).

This receptor is known to be a strong luteolytic mediator, and it is thought to be involved in modulating intraocular pressure and smooth muscle contraction in uterus.

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Assay Characterization

Our expression plasmid contains the coding sequence of human PTGFR protein. Our plasmid was transfected in U2OS cells. Resistant clones were obtained by limit dilution and receptor gene expression was tested by RT-PCR using GAPDH as internal control (Fig.1).

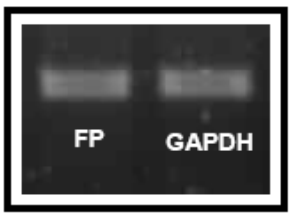

Fig.1. PTGFR and GAPDH housekeeping gene RT-PCR.

Validation of PTGFR cell line

Calcium assay (Ec50 = 3.29 x 10^{-8}M)

A typical fluorescent calcium assay was performed using Fura-2/AM ratiometric. Calcium increase inside the cell was measured using the ratio of the fluorescence from Fura2 bound and not bound to the ion. Image acquisition was performed using a "BD Pathway 855" High-Content Bioimager from BD Biosciences.

Cells were incubated with Fura2-AM and treated with increasing PGE2 concentrations.

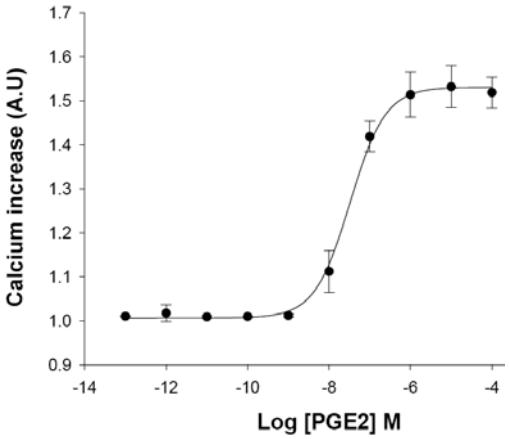

Fig.2. PTGFR dose response in calcium assay. Cells were treated with PGE2 concentrations ranging from 0 to 100 µM, n=6. The EC50 for PGE2 was ~3.29x10^{-8}M. The calcium assay was validated with a Z´= 0.78+/- 0.02 for High Throughput Screening.

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