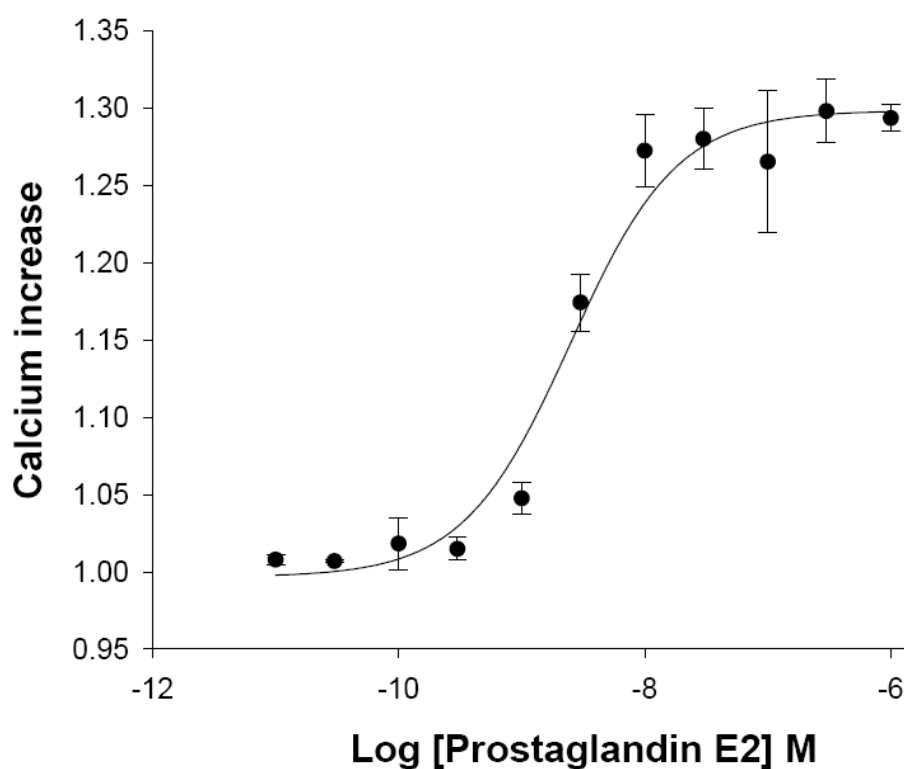


## HiTSeeker CELL LINES (LABEL-FREE GPCRS)

### - PROSTAGLANDIN E RECEPTOR 1, SUBTYPE EP1 (PTGER1)(EP1) CELL LINE -



**Product name:** PTGER1 (EP1) /U2OS cell line

**EC<sub>50</sub> Prostaglandin E2:**  $2.48 \times 10^{-9}$  M

**Z':** 0.88+/- 0.02

## - PROSTAGLANDIN E RECEPTOR 1, SUBTYPE EP1 (PTGER1) U2OS CELL LINE -

<b>Product Name:</b>	PTGER1 (EP1)/U2OS
<b>Official Full Name:</b>	Prostaglandin E receptor 1, subtype E1
<b>DNA Accession Number:</b>	GenBank: AY275470
<b>Host Cell:</b>	U2OS
<b>Format:</b>	2 cryopreserved vials
<b>Resistance:</b>	Puromycin
<b>Size:</b>	<i>P30402</i> : 2 vials of $3 \times 10^6$ proliferative cells <i>P30402-DA</i> : 1 vial of $2.5 \times 10^6$ division-arrested cells
<b>Storage:</b>	Liquid Nitrogen

### **Assay Briefly description**

Each vial of HiTSeeker PTGER1 contains U2OS cells stably expressing human Prostaglandin E1 receptor subtype E1 (PTGER1) with no tag.

Innoprot's HiTSeeker PTGER1 cell line has been designed to assay compounds or analyze their capability to modulate Prostaglandin E1 receptor subtype E1. When the agonist binds to PTGER1 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 PTGER1 activation process in High Throughput Screening.

### **About PTGER1**

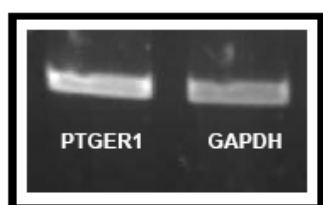
Prostaglandins are a group of lipid compounds that participate in a wide range of body functions.

This protein, which belongs to the G protein-coupled receptor family, is one of four receptors identified for prostaglandin E2 (PGE2).

Some studies suggest a role of this receptor in mediating algia and in regulation of blood pressure and it has also been suggested that this gene may mediate adrenocorticotrophic hormone response to bacterial endotoxin.

## **Assay Characterization**

Our expression plasmid contains the coding sequence of human PTGER1 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).



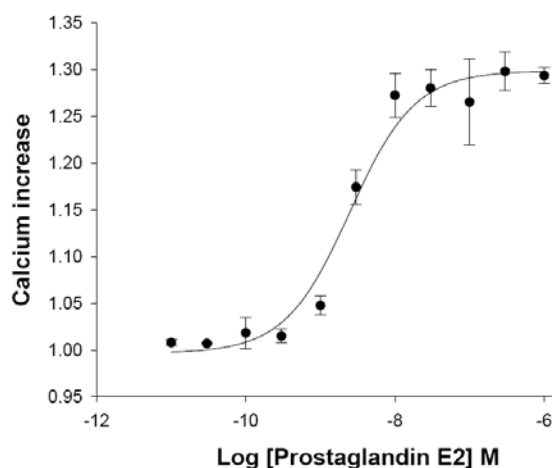
**Fig.1.** PTGER1 and GAPDH housekeeping gene RT-PCR.

## **Validation of PTGER1 cell line**

### **Calcium assay ( $EC_{50} = 2.48 \times 10^{-9} M$ )**

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 Prostaglandin E2 concentrations.



**Fig.2. PTGER1 dose response in calcium assay.** Cells were treated with Prostaglandin E2 concentrations ranging from 0 to 1  $\mu M$ ,  $n=5$ . The  $EC_{50}$  for Prostaglandin E2 was  $\sim 2.48 \times 10^{-9} M$ . The calcium assay was validated with a  $Z' = 0.88 \pm 0.02$  for High Content Screening.