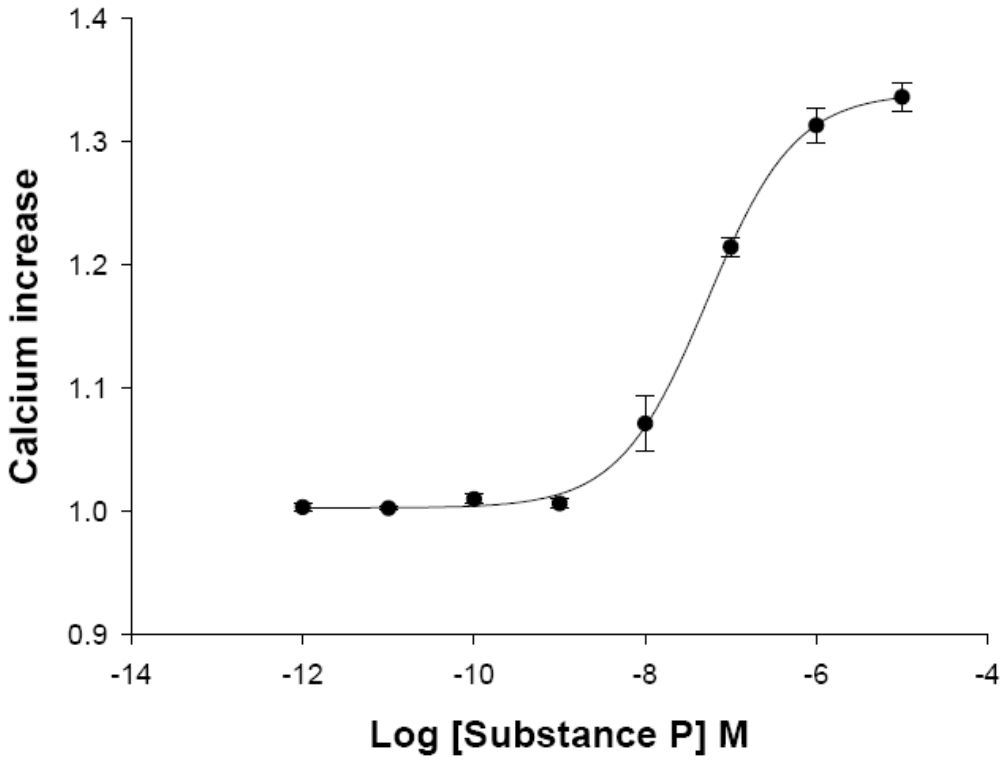
REF: P30149

HiTSeeker CELL LINES (LABEL-FREE GPCRS)

-TACHYKININ RECEPTOR 3 CELL LINE -



Product name: TACR3 (NK3) /U2OS cell line

Ec_{50} Substance P: 5.3x 10^{-8} M

Z´: 0.86+/- 0.02

日本総代理店
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- TACHYKININ RECEPTOR 3 U2OS CELL LINE -

Product Name:	TACR3 (NK3)/U2OS
Official Full Name:	Tachykinin receptor 3
DNA Accesion Number:	GenBank: AY462099
Host Cell:	U2OS
Format:	2 cryopreserved vials
Resistance:	G418
Size:	*P30149*: 2 vials of 3 x 10^6 proliferative cells *P30149-DA*: 1 vial of 2.5x10^6 division-arrested cells
Storage:	Liquid Nitrogen

Assay Briefly description

TACR3/U2OS contains U2OS cells stably expressing human Tachykinin receptor 3 (TACR3) with no tag.

Innoprot TACR3 cell line has been designed to assay compounds or analyze their capability to modulate Tachykinin receptor 3. When the agonist binds to TACR3 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 TACR3 activation process in High Throughput Screening.

About TACR3

Tachykinin receptor 3 is the gene that encodes a protein that is one of the three Tachykinin receptors (TACRs), also termed NKRs.

The Tachykinin receptor family is a group of G-coupled receptors whose principal ligands are the Neurokinins.

TACR3 is distributed throughout the CNS and it is found in high levels in the cerebral cortex, basal ganglia and dorsal horn of the spinal cord.

TACR3 ligands are used in clinical trials for treatment of schizophrenia and other indications.

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Assay Characterization

Our expression plasmid contains the coding sequence of human TACR3 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).

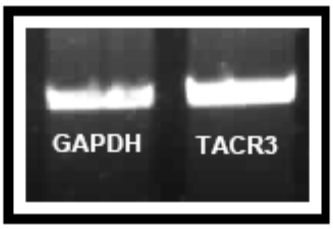

Fig.1. GAPDH housekeeping gene and TACR3 RT-PCR.

Validation of TACR3 *cell line*

Calcium assay (Ec50 = 5.3 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 Substance P concentrations.

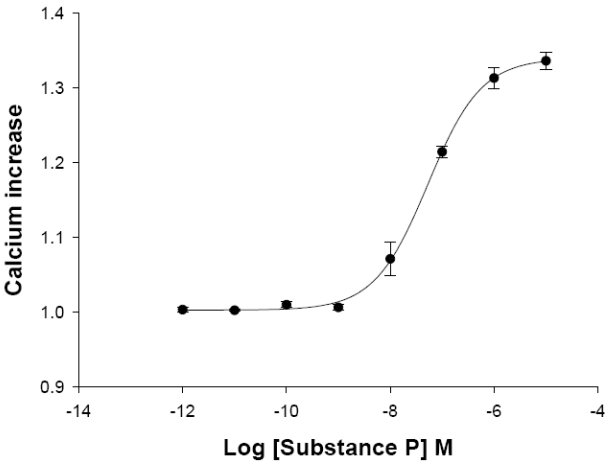

Fig.2. TACR3 dose response in calcium assay.
Cells were treated with **Substance P** concentrations ranging from 0 to 10 µM, n=5. The EC50 for **Substance P** was ~**$5.3x10^{-8}$M**. The calcium assay was validated with a Z´= 0.86+/- 0.02 for High Content Screening.

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