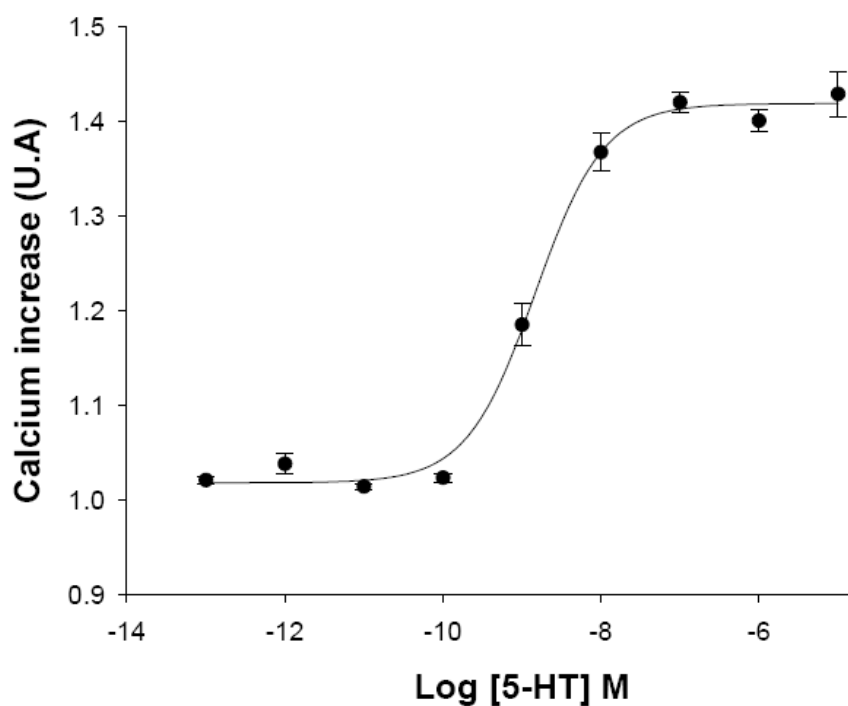


HiTSeeker CELL LINES (LABEL-FREE GPCRS)

- 5-HYDROXYTRYPTAMINE RECEPTOR 2C (HTR2C) CELL LINE -



Product name: HTR2C (5-HT_{2C}) /U2OS cell line

EC₅₀ 5-HT: 1.16 x 10⁻⁹ M

Z': 0.80+/- 0.02

- 5-HYDROXYTRYPTAMINE RECEPTOR 2C (HTR2C) U2OS CELL LINE -

Product Name:	HTR2C (5-HT _{2C})/U2OS
Official Full Name:	5-Hydroxytryptamine receptor 2C
DNA Accession Number:	GenBank: NM_000868
Host Cell:	U2OS
Format:	2 cryopreserved vials
Resistance:	G418
References:	 P30162: 2 vials of 3 x 10⁶ proliferative cells  P30162-DA: 1 vial of 2 x 10⁶ division-arrested cells
Storage:	Liquid Nitrogen

Assay Briefly description

Each vial of HiTSeeker HTR2C contains U2OS cells stably expressing human 5-Hydroxytryptamine receptor 2C (HTR2C) with no tag.

Innoprot's HiTSeeker HTR2C cell line has been designed to assay compounds or analyze their capability to modulate 5-Hydroxytryptamine receptor 2C. When the agonist binds to HTR2C 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 HTR2C activation process in High Throughput Screening.

About HTR2C

The **serotonin receptors**, also known as **5-hydroxytryptamine receptors** or **5-HT receptors** are a group of G protein-coupled receptors (GPCRs) found in the central and peripheral nervous systems. They mediate both excitatory and inhibitory neurotransmission. The neurotransmitter serotonin acts as their natural ligand.

HTR2C receptor plays an important role mediating the interaction between serotonergic and dopaminergic systems, that is why they have been involved in the regulation of mood, affective behaviour, and memory.

Assay Characterization

Our expression plasmid contains the coding sequence of human HTR2C 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).

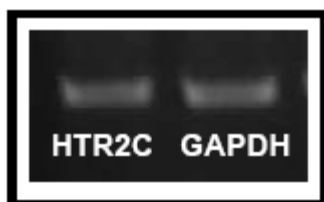


Fig.1. HTR2C and GAPDH housekeeping gene RT-PCR.

Validation of HTR2C cell line

Calcium assay ($EC_{50} = 1.16 \times 10^{-9} M$)

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 5-HT concentrations.

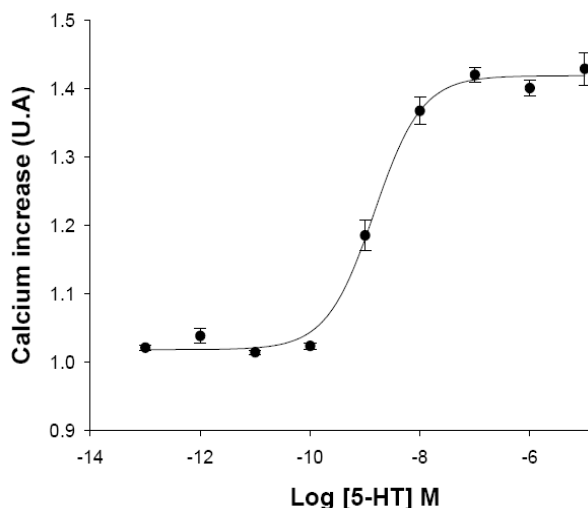


Fig.2. HTR2C dose response in calcium assay.

Cells were treated with **5-HT** concentrations ranging from 0 to 10 μM , $n=5$. The EC_{50} for **5-HT** was $\sim 1.16 \times 10^{-9} M$. The calcium assay was validated with a $Z' = 0.80 \pm 0.02$ for High Throughput Screening.