NOMAD CELL LINES

P70410-G

Nomad Biosensors™ comprises a family of genetically encoded fluorescent sensors designed to monitor the signaling of G protein-coupled receptors (GPCRs) in cell-based assays.

Nomad Biosensors are engineered to measure the intracellular dynamics of second messengers such as calcium (Ca^{2+}Nomad), cAMP (cAMPNomad) or diacylglycerol (DAGNomad) once the GPCRs are activated. Additionally, the β-arrestin signaling can also be studied with the ArresNomad biosensors. Actually, Nomad biosensors can be combined in the same cell line for multiplex assays.

Prior to GPCR activation, Nomad biosensors are localized in the plasma membrane. Upon ligand binding, the sensors undergo a conformational change that leads to an increase in the fluorescence intensity and their relocalization in the vesicular trafficking of the cells.

Innoprot

Ca^{2+}NOMAD M_1 Cell Line

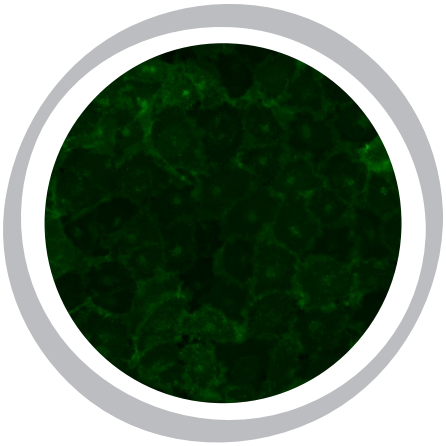
Muscarinic Acetylcholine Receptor M_1

Ca^{2+} Nomad Biosensor

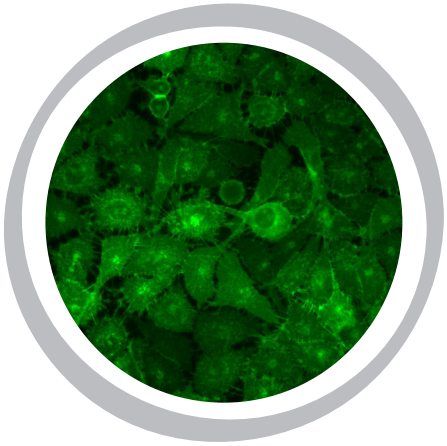
Nomad cell lines stably express both a biosensor and a GPCR (untagged). The activity of the receptor can be easily quantified on living cells by image analysis in a High Content Screening (HCS) assay or by fluorescence intensity emission in a High Throughput Screening (HTS) assay.

Innoprot´s Nomad cell lines have been designed to assay compounds or analyze their capability to modulate the signaling of the GPCRs.

Each vial of Ca^{2+}Nomad-M1 contains U2OS cells stably expressing the green Ca^{2+}Nomad biosensor and the cholinergic receptor muscarinic 1. When an agonist binds to M1 a G protein is activated, which in turn, triggers a cellular response mediated by calcium. This cell line has been validated measuring the fluorescence intensity emission of the green Ca^{2+}Nomad biosensor within the cell.



Control



Oxotremorine

Product Name: Ca^{2+}Nomad-M1 cell line
Reference: P70410-G
Recp. Official Full Name: Cholinergic receptor muscarinic 1 receptor
DNA Accession Number: BC007740
Host Cell: U2OS
Resistance: G418 + Puromycin
Quantity: > 3 x 10^6 cells / vial
Storage: Liquid Nitrogen

NOMAD CELL LINES

Calcium assay

The green Ca^{2+}Nomad-M1 cell line was plated in a 96 well-plate and incubated for a minimum of 4 hours and not more than 24 hours at 37 °C and 5% CO_2 to let the cells attach to the plate surface. Then, cells were treated with oxotremorine diluted in a serum-reduced medium during 20-24 hours.

The increase in the fluorescence intensity of the green Ca^{2+}Nomad biosensor was detected and analyzed using a conventional microplate reader. Images were captured with an image analysis equipment.

Ca^{2+}Nomad-M1 (U2OS cell line)

Ec_{50} Ca^{2+} assay: 9.17×10^{-7} M | Z´Ca^{2+}: 0.81

Agonism Assay

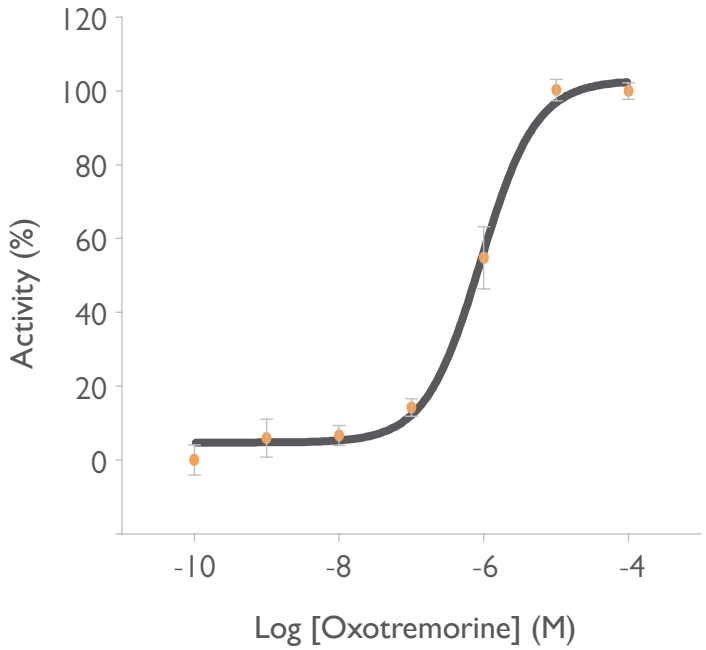


Figure 1. Agonism dose-response curve. Cells were stimulated with increasing dilutions of oxotremorine overnight. Data were normalized as percentages of activity of the green Ca2+Nomad biosensor against the positive control (Oxotremorine 100 µM) after subtracting the value of the vehicle control.

INNOVATIVE TECHNOLOGIES IN BIOLOGICAL SYSTEMS, S.L.
Parque Tecnológico Bizkaia, Edifício 502, 1ª Planta | 48160 | Derio | Bizkaia
Tel.: +34 944005355 | Fax: +34 946579925
innoprot@innoprot.com | www.innoprot.com

Innoprot