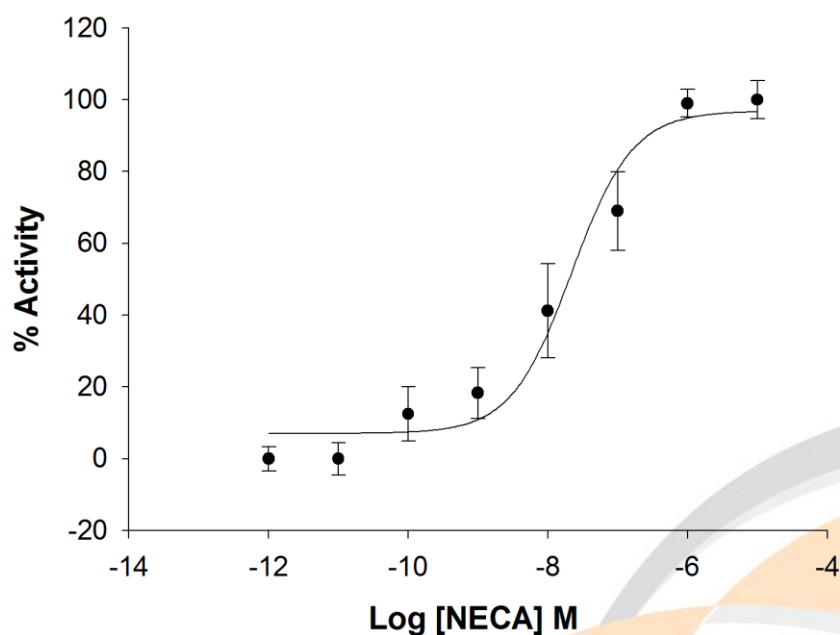
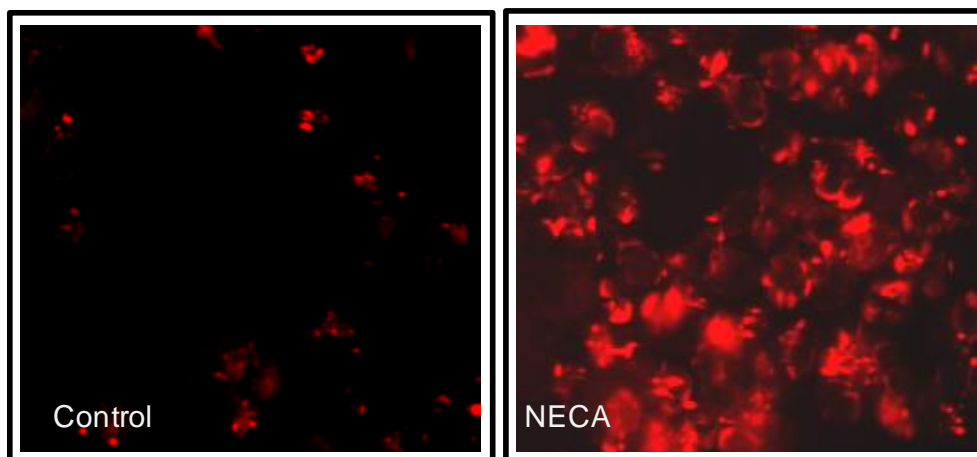


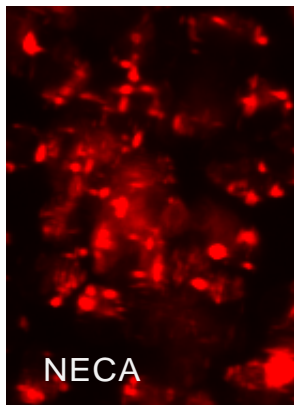
cAMP NOMAD-FP650 cell lines -ADENOSINE A1 RECEPTOR (ADORA1)-



Red _{cAMP}Nomad-ADORA1 (HEK293 cell line)

EC₅₀ NECA: 2.19x10⁻⁸ M

Z': 0.74+/- 0.01



Product Name: ADORA1_{cAMP}Nomad cell line

Reference: P70523

Recp. Official Full Name: Adenosine A1 receptor

DNA Accession Number: NM_000674

Host Cell: HEK293

Resistance: G418 + Hygromycin

Quantity: > 3 x 10⁶ cells / vial

Storage: Liquid Nitrogen

Assay Briefly description

Each vial of _{cAMP}Nomad ADORA1 contains HEK293 cells stably expressing _{cAMP}Nomad-FP650 biosensor and adenosine A1 receptor (with no tag).

Innoprot _{cAMP}Nomad ADORA1 cell line has been designed to assay compounds or analyze their capability to modulate Adenosine A1 receptor. When an agonist binds to ADORA1 a G protein is activated, which in turn, triggers a cellular response mediated by cAMP. This cell line has been validated measuring cAMP decrease in the cytosol analyzing _{cAMP}Nomad biosensor distribution within the cell. This cell line allows the image analysis of the stimuli induced by the compounds.

This highly reproducible assay has been validated using NECA as agonist in a High Throughput Analysis (HTA).

About Red _{cAMP}Nomad Biosensor

Red _{cAMP}Nomad Biosensor is a fluorescent polypeptide that in the presence or absence of cAMP changes its localization within the cell.

Before cAMP production stimulation, the fluorescent biosensor is localized in the cellular membrane. An increase/decrease in this second messenger concentration leads to a change in the structural folding of red _{cAMP}Nomad Biosensor promoting its cellular relocation in the vesicular trafficking of the cells.

In a cell line co-expressing red _{cAMP}Nomad Biosensor and a GPCR of interest, the activity can be easily quantified on living cells by image analysis of fluorescence granularity or fluorescence intensity analysis.

 **cAMP Assay**

Red_{cAMP}Nomad HEK293 cells, stably expressing adenosine A1 receptor (ADORA1), were stimulated with 8 log dilution series ranging from 0 to 10 μ M of NECA during 24h (n=5). % Activity was calculated relative to positive (10 μ M).

Fluorescence intensity analysis

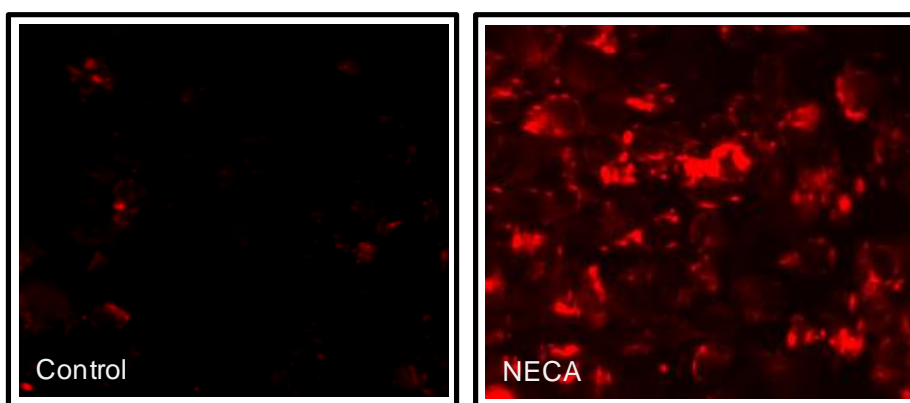


Fig1. Red_{cAMP}Nomad biosensor negative control and NECA stimulation.

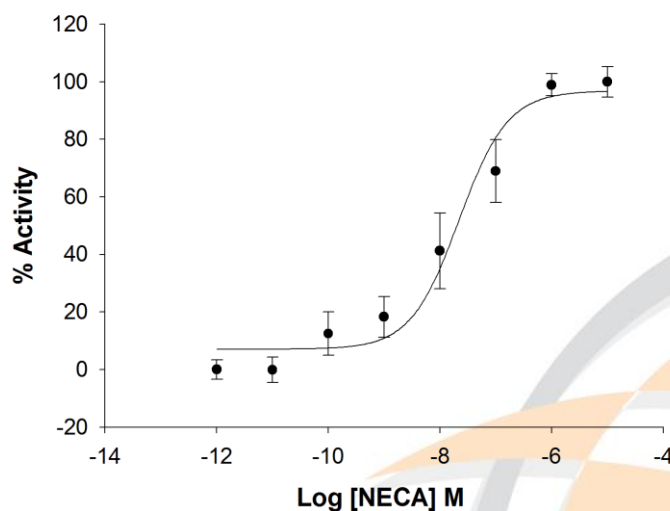


Fig 2. Concentration-response curve for adenosine A1 receptor in Red_{cAMP}Nomad-ADORA1 cell line analyzed using “Synergy 2” microplate reader from Biotek. The EC₅₀ for NECA was $2,19 \times 10^{-8}$ M after a treatment of 24 h with the agonist. The assay was validated with an average of $Z' = 0.74 \pm 0.01$.