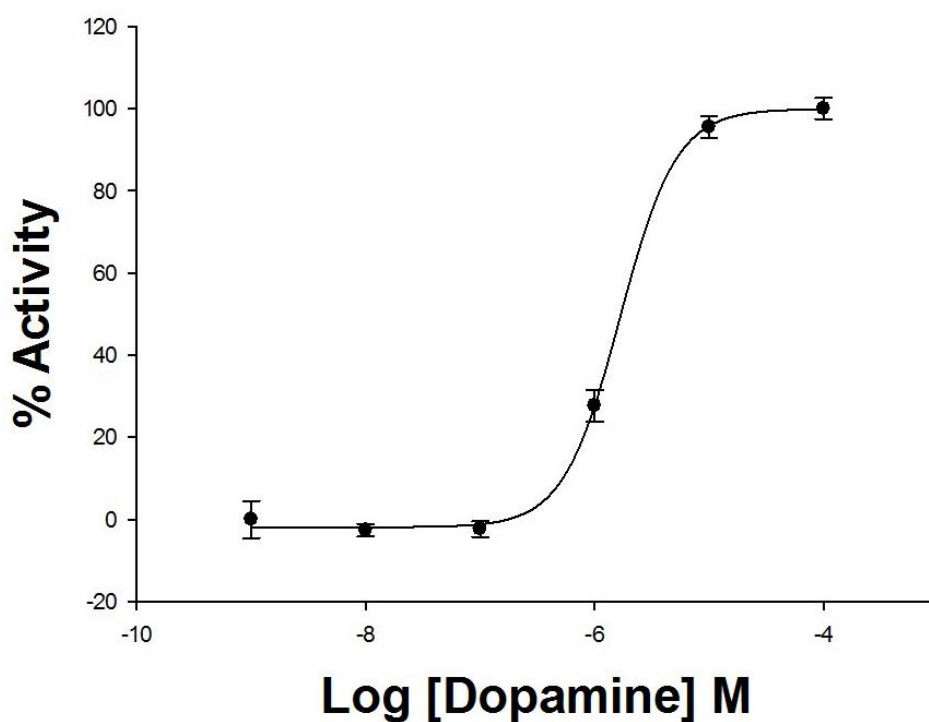
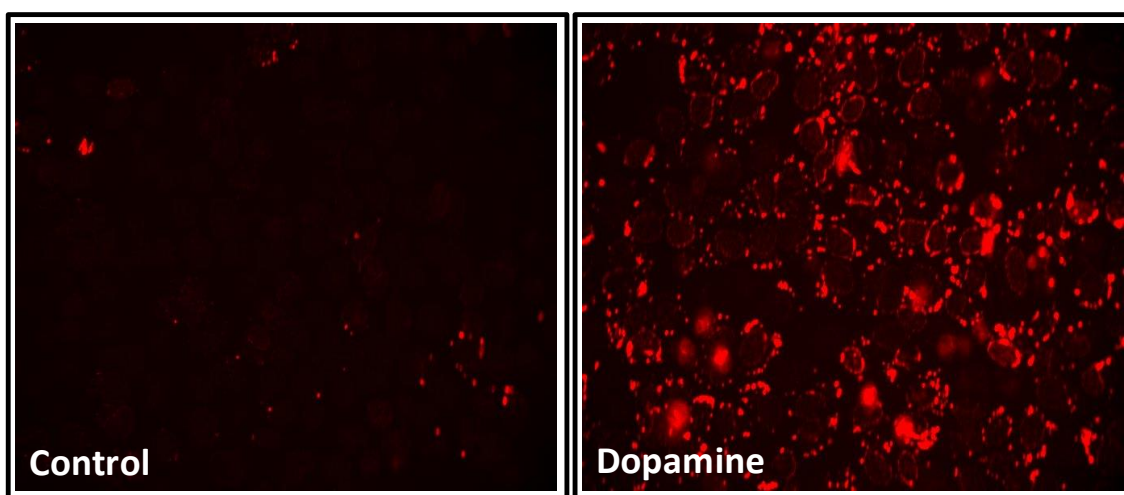


## cAMP NOMAD-FP650 CELL LINES

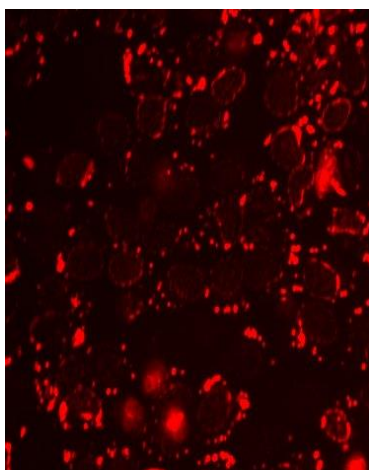
### -DOPAMINE RECEPTOR D5 (DRD5)-



Red cAMPNomad-DRD5 (U2OS cell line)

EC<sub>50</sub> Dopamine:  $1.66 \times 10^{-6}$  M

Z': 0.79



**Product Name:** DRD5 <sub>cAMP</sub>Nomad cell line  
**Reference:** P70509  
**Recep. Official Full Name:** Dopamine receptor D5  
**DNA Accession Number:** NC\_000004.12  
**Host Cell:** U2OS  
**Resistance:** G418 + Puromycin  
**Quantity:** > 3 x 10<sup>6</sup> cells / vial  
**Storage:** Liquid Nitrogen

### Assay Briefly description

Each vial of red <sub>cAMP</sub>Nomad-DRD5 contains U2OS cells stably expressing red <sub>cAMP</sub>Nomad biosensor and Dopamine receptor D5 (with no tag).

Innopro's red <sub>cAMP</sub>Nomad-DRD5 cell line has been designed to assay compounds or analyze their capability to modulate Dopamine receptor D5. When an agonist binds to DRD5 a G protein is activated, which in turn, triggers a cellular response mediated by cAMP.

This cell line has been validated measuring cAMP increase in the cytosol analyzing red <sub>cAMP</sub>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 Dopamine as agonist in a High Content Analysis (HCA) and a High Throughput Analysis (HTA).

### About Red <sub>cAMP</sub>Nomad Biosensor

Red <sub>cAMP</sub>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 in this second messenger concentration leads to a change in the structural folding of red <sub>cAMP</sub>Nomad Biosensor promoting its cellular relocation in the vesicular trafficking of the cells.

In a cell line co-expressing red <sub>cAMP</sub>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.

cAMPNomad U2OS cells, stably expressing Dopamine receptor D5 (DRD5), were stimulated with 9 log dilution series ranging from 0 to 100  $\mu$ M of Oxotremorine during 24h (n=4). %Activity was calculated relative to positive (100  $\mu$ M).

### Fluorescence intensity analysis

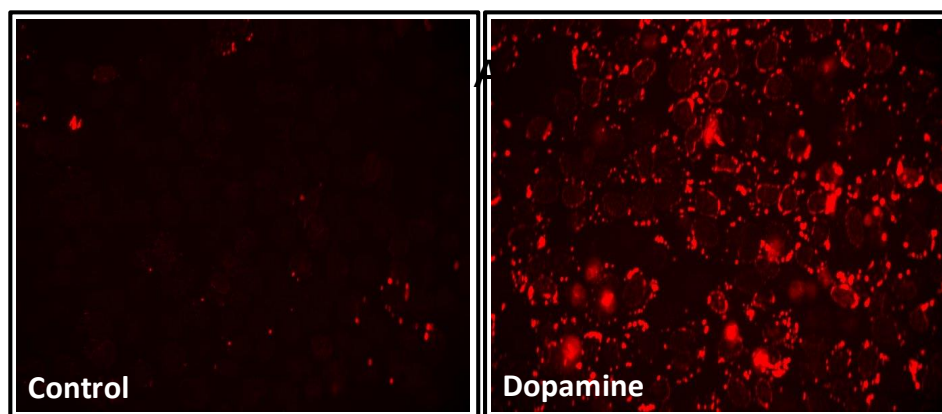


Fig1. Red cAMPNomad biosensor negative control and Dopamine stimulation.

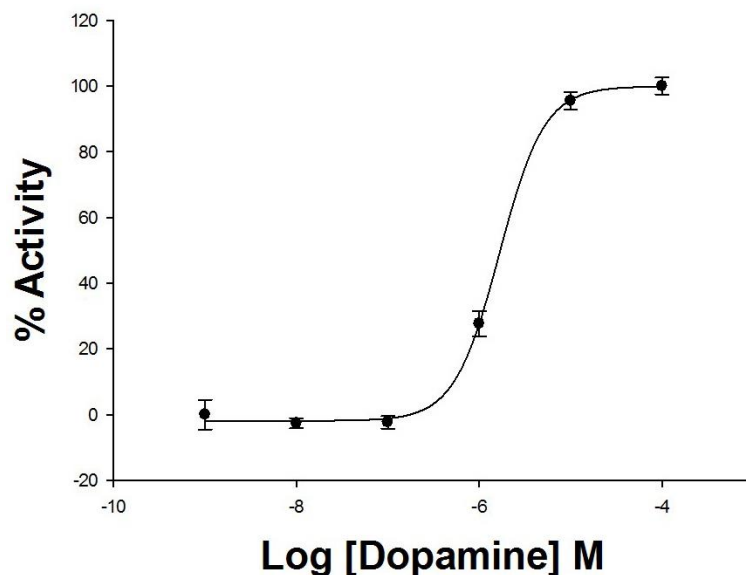


Fig 2. Concentration-response curve for Dopamine in Red cAMPNomad-TSHR cell line analyzed using “Synergy 2” microplate reader from Biotek. The EC<sub>50</sub> for Dopamine was  $1.66 \times 10^{-6}$  M after a treatment of 24 h with the agonist. The assay was validated with an average of  $Z' = 0.79$ .