Metabotropic Glutamate Receptor 7 (GRM7) ACTOne™ Stable Cell Line

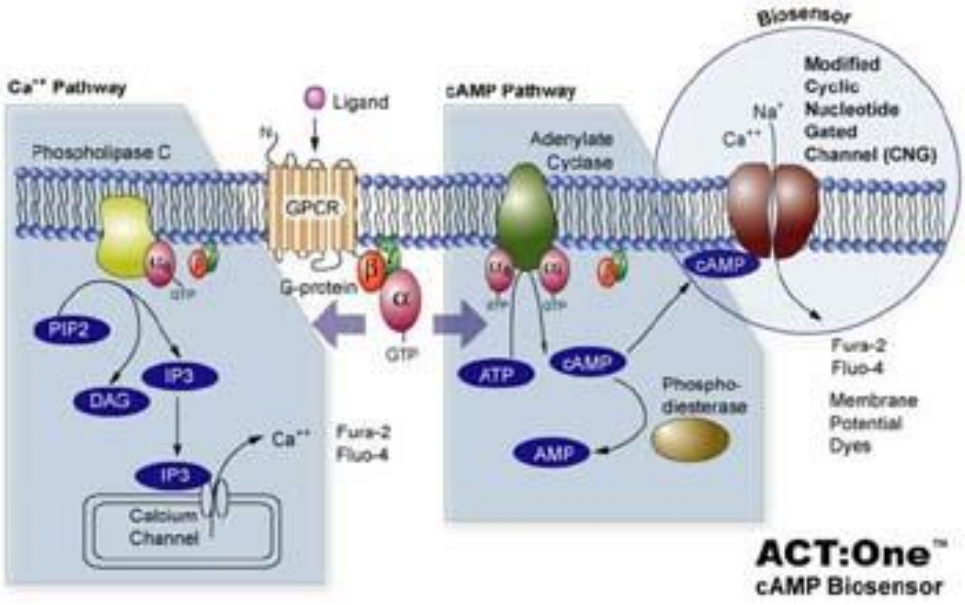
CATALOG NUMBER: CL-11-GRM7

Introduction

The metabotropic glutamate receptors are a family of G protein-coupled receptors, that have been divided into 3 groups on the basis of sequence homology, putative signal transduction mechanisms, and pharmacologic properties. Group I includes GRM1 and GRM5 and these receptors have been shown to activate phospholipase C. Group II includes GRM2 and GRM3 while Group III includes GRM4, GRM6, GRM7 and GRM8. Group II and III receptors are linked to the inhibition of the cyclic AMP cascade but differ in their agonist selectivity.

Description

Human GRM7 ACTOne™ is a HEK-293 CNG-Slcla3 cell line that expresses recombinant human GRM7. HEK-293 CNG-Slcla3 cells express a modified CNG (Cyclic Nucleotide Gated) channel that opens in response to elevated intracellular cAMP levels and consequently result in ion flux (often detectable by calcium-responsive dye, Cat# CA-C155) and cell membrane depolarization which can be easily measured with fluorescent Membrane Potential Dye (Cat# CA-M165). The assay allows both end-point and kinetic measurement of intracellular cAMP changes with a FLIPR, or a fluorescence microplate reader.



Parental Cells

HEK-293 CNG-Slcla3 cells (Cat# CL-03-PC40)

Gene/Enzyme Introduced

GRM7 (Genbank Accession No. AAB51763.1)

Applications

- cAMP dependent assay for Gi-coupled human GRM7
- cell based high-throughput screening of human GRM7 inhibitors

Functional Test

- this cell line has been tested positive for GRM7 specific response

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- surviving rate: More than 2.5 million/vial on the second day after thawing
- the receptor specific activity is stable for 10 weeks continuous passage

Mycoplasma Contamination Test

This lot of cells has been tested and found to be free of mycoplasma contamination.

Content

- Stable cells: 1 mL (1 x 10^6 cells/mL in 70% DMEM, 20% FBS, 10% DMSO)

Growth Properties

Adherent

Cell Culture Medium

- Growth medium: DMEM-10% FBS supplemented with 250 µg/ml G418, 1 µg/ml Puromycin
- Freezing medium: 10% DMSO, 90% complete cell culture medium

Storage

Remove the frozen cells from the dry ice packaging and immediately place the cells at a temperature below -130°C, preferably in liquid nitrogen vapor, until ready for use.

Data Analysis

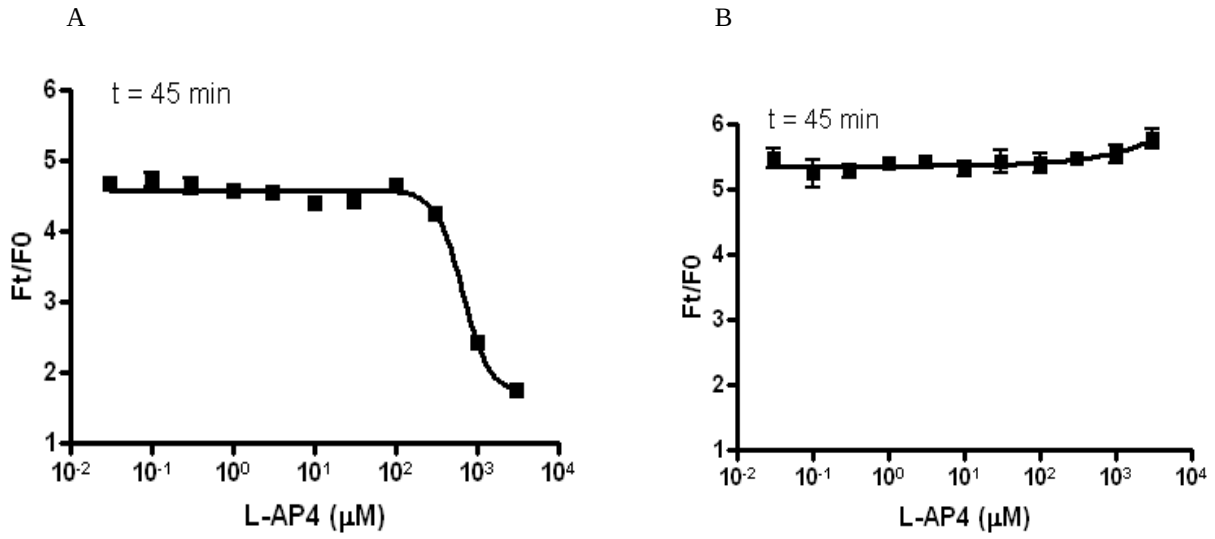

Figure 1. Response of ACT*One*™ GRM7 cell line & parental cell line to L-(+)-2-Amino-4-phosphonobutyric acid.

ACTOne™ GRM7 cells and parental cells (Cat# CL-03-PC20) were plated overnight in 20 µl culture medium on a 384 well Biocoat plate. The next day, cells were dye-loaded with 20 µl/well of 1x Dye-loading solution (membrane potential dye kit, Cat# CA-M165). After 2 hours of incubation at room temperature, two readings were obtained prior to and 30 min after the addition of L-(+)-2-Amino-4-phosphonobutyric acid. Ratios of the two readings (F/F0) are plotted in the figure.

A. Dose response curve of L-(+)-2-Amino-4-phosphonobutyric acid in ACT*One*™ GRM7 cell line. EC50 = 657 µM in the presence of PDE inhibitor Ro 20-1724 and β–adrenoceptor agonist isoproterenol.

B. Parental cells do not respond to L-(+)-2-Amino-4-phosphonobutyric acid.

Notice to Purchaser

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