

## Sphingosine-1-Phosphate Receptor 3 (S1PR3) ACTOne™ Stable Cell Line

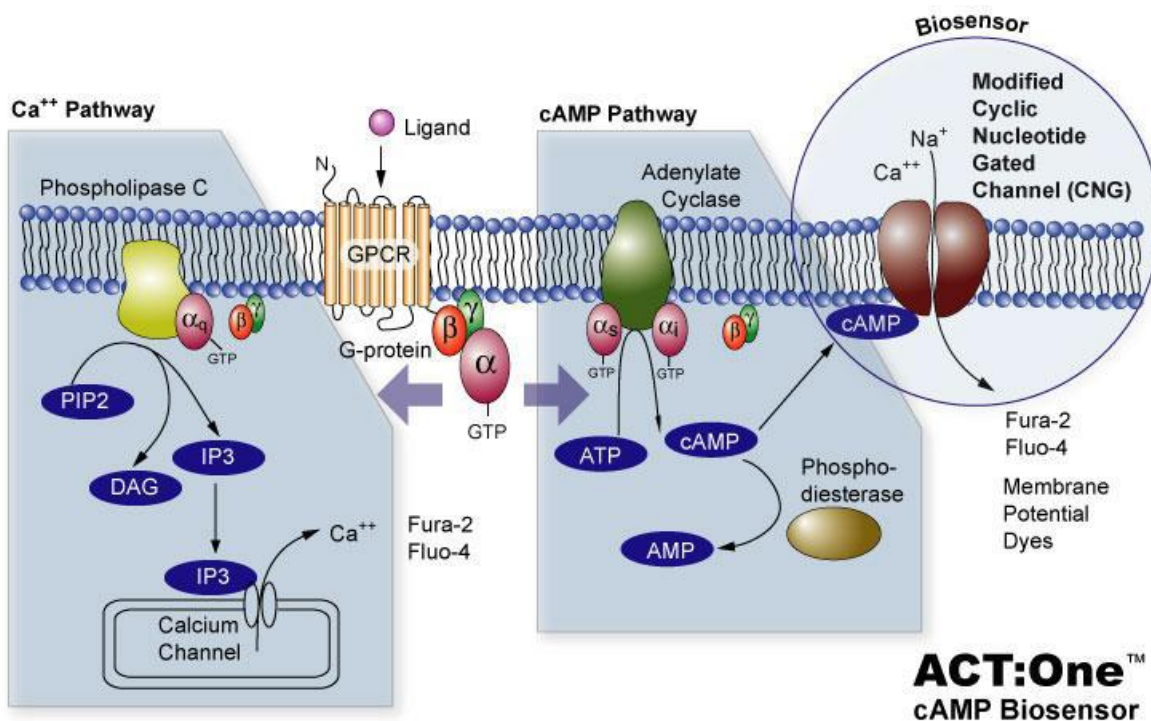
CATALOG NUMBER: CL-01-S1PR3

### Introduction

S1PR3 is a G-protein-coupled receptor which binds the bioactive signaling molecule sphingosine 1-phosphate (S1P). S1PR3 belongs to a sphingosine-1-phosphate receptor subfamily comprising five members (S1PR3-5). They are expressed in a wide variety of tissues, with each subtype exhibiting a different cell specificity, although they are found at their highest density on leukocytes. S1PR3 has been identified as a functional receptor for sphingosine 1-phosphate and likely contributes to the regulation of angiogenesis and vascular endothelial cell function.

### Description

Human S1PR3 ACTOne™ is a HEK-293 CNG cell line that expresses recombinant human S1PR3. HEK-293 CNG 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 FDSS, FLIPR, or a fluorescence microplate reader.



### Parental Cells

HEK-293 CNG cells (originally developed by BD Biosciences by introducing CNG in HEK-293 cells) (Cat# CL-03-PC20)

### Gene/Enzyme Introduced

S1PR3 (Genbank Accession No. NP\_005217.2)

### Applications

- cAMP dependent human S1PR3 receptor cell based assay

- cell based high-throughput screening of human S1PR3 receptor agonists/antagonists

## Functional Test

- this cell line has been tested positive for S1PR3 receptor specific response
- 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 S1PR3 receptor cells: 1 mL (1 x 10<sup>6</sup> cells/mL in 70% DMEM, 20% FBS, 10% DMSO)

## Growth Properties

Adherent

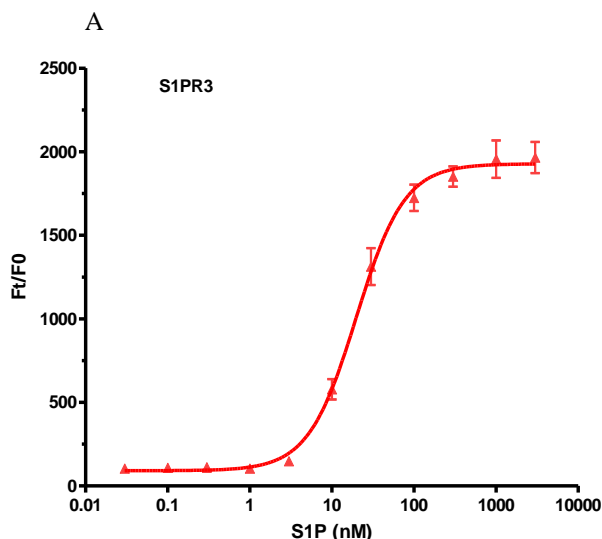
## Cell Culture Medium

- 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 Example



### Figure 1. Response of ACTOne™ S1PR3 cell line

ACTOne™ S1PR3 receptor 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 (Elite™ Non-Wash Calcium Dye, Cat# CA-C155). After 1 hour of incubation at 37 °C, the plate was read on Hmamastu FDSS 7000 with online addition of the ligand. Fluorescent intensity was used to plot the figure.

- A. Dose response curve of S1P in ACTOne™ S1PR3 cell line. EC50 = 20 nM**  
 \*Parental cells do not respond to S1P (data not shown)



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