

Secretin Receptor (SCTR) ACTOne™ Stable Cell Line

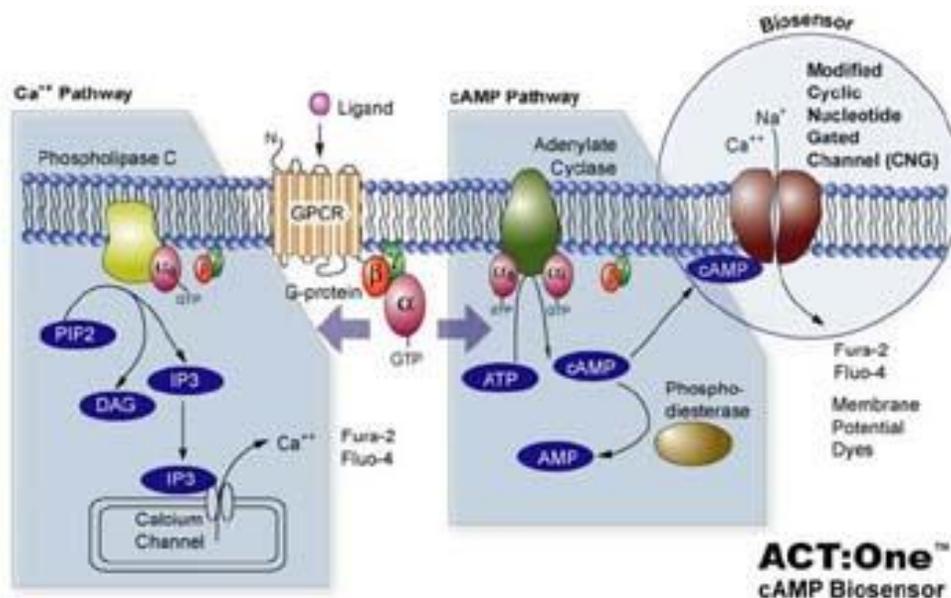
CATALOG NUMBER: CL-01-SCTR

Introduction

SCTR is a G protein-coupled receptor and belongs to the glucagon-VIP-secretin receptor family. It binds secretin which is the most potent regulator of pancreatic bicarbonate, electrolyte and volume secretion. Secretin and its receptor are suggested to be involved in pancreatic cancer and autism.

Description

Human SCTR ACTOne™ is a HEK-293 CNG cell line that expresses recombinant human SCTR. 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 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

SCTR (Genbank Accession No. AAR25625)

Applications

- cAMP dependent human SCTR cell based assay
- cell based high-throughput screening of human SCTR inhibitors

Functional Test

- this cell line has been tested positive for SCTR 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 cells: 1 mL (1 x 10⁶ 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

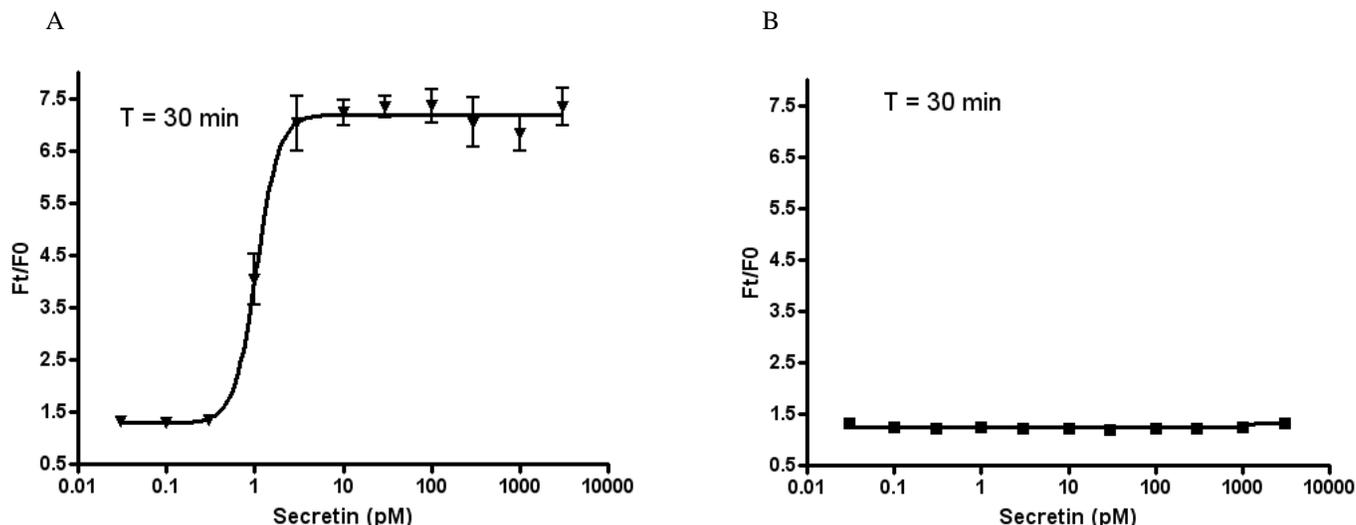


Figure 1. Response of ACTOne™ SCTR cell line & parental cell line to secretin

ACTOne™ SCTR 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 secretin. Ratios of the two readings (F/F0) are plotted in the figure.

- Dose response curve of secretin in ACTOne™ SCTR cell line. EC50 = 1.04 pM in the presence of PDE inhibitor Ro 20-1724, and EC50 = 3.17 pM in the absence of Ro20-1724 (data not shown).
- Parental cells do not respond to secretin.

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