Human Recombinant SST1 Somatostatin Receptor Stable Cell Line

Cat. No. M00473 **Version 06092014**

I. INTRODUCTION

Catalog Number: M00473
Cell Line Name: CHO-K1/SST1/Gqi5
Gene Synonyms: SSTR1
Expressed Gene: Genbank Accession Number NM_001049; no expressed tags
Host Cell: CHO-K1/Gqi5
Quantity: Two vials of frozen cells (3×10^6 per vial)
Stability: 16 passages
Application: Functional assay for SST1 receptor
Freeze Medium: 45% culture medium, 45% FBS, 10% DMSO
Complete Culture Medium: Ham's F12, 10% FBS
Culture Medium: Ham's F12, 10% FBS, 200 μg/ml Zeocin, 100 μg/ml Hygromycin B
Mycoplasma Status: Negative
Storage: Liquid nitrogen immediately upon delivery

II. BACKGROUND

Somatostatin acts at many sites to inhibit the release of many hormones and other secretory proteins. The biologic effects of somatostatin are probably mediated by a family of G protein-coupled receptors that are expressed in a tissue-specific manner. SST1 receptor is a G_i/G_o-coupled GPCR which is expressed in pancreatic islets, pituitary, Cerebellum (Purkinje cells), frontal cortex (pyramidal cells), hippocampus (CA1-4 subfields and some granule cells of the dentate gyrus). It inhibits cAMP accumulation and stimulates tyrosine phosphatase activity, it also can antiproliferation.

§: GenScript employs a PCR-based method to test the mycoplasma. The test covers 11 of the most common strains of mycoplasma, (covering approximately 95% of M. fermentans, M. hyorhinis, M. arginini, M. orale, M. salivarium, M. hominis, M. pulmonis, M. arthritidis, M. neurolyticum, M. hyopneumoniae and M. capricolum) and one species Ureaplasma (U. urealyticum), with sufficient sensitivity and specificity.

III. REPRESENTATIVE DATA

Concentration-dependent stimulation of intracellular calcium mobilization by Somatostatin-14 in CHO-K1/SST1/Gα15 and CHO-K1/Gα15 cells

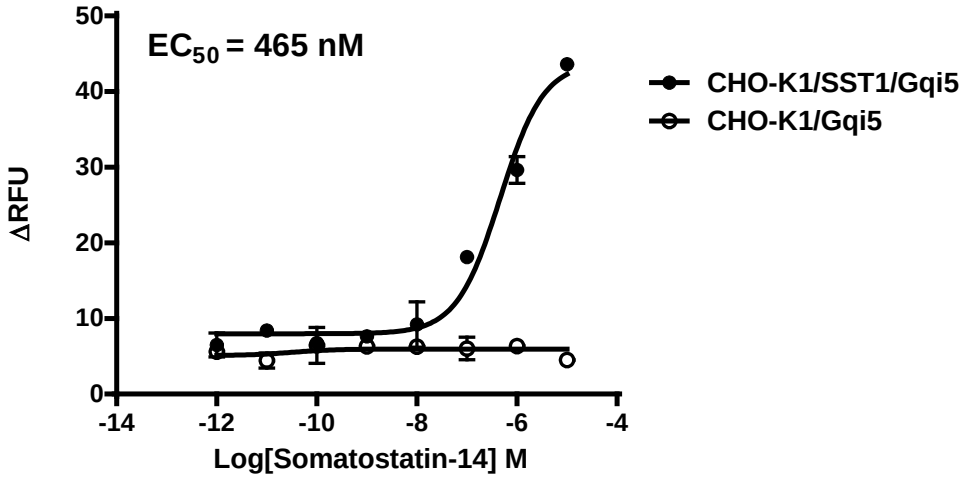

Figure 1. Somatostatin-14-induced concentration-dependent stimulation of intracellular calcium mobilization in CHO-K1/SST1/Gα15 and CHO-K1/Gα15 cells. The cells were loaded with Calcium-4 prior to stimulation with an SST1 receptor agonist, Somatostatin-14. The intracellular calcium change was measured by FlexStation. The relative fluorescent units (RFU) were plotted against the log of the cumulative doses (10-fold dilution) of Somatostatin-14 (Mean ± SD, n = 2). The EC_{50} of Somatostatin-14 on SST1 co-expressing with Gα15 in CHO-K1 cells was 465 nM. The S/B of Somatostatin-14 on SST1 co-expressing with Gα15 in CHO-K1 cells was 33.

Notes:

1. EC_{50} value is calculated with four parameter logistic equation:
   Y=Bottom + (Top-Bottom)/(1+10^((LogEC_{50}-X)*HillSlope))
   X is the logarithm of concentration.
   Y is RFU and starts at Bottom and goes to Top with a sigmoid shape.
2. Signal to background Ratio (S/B) = Top/Bottom

IV. THAWING AND SUBCULTURING

Thawing Protocol

1. Remove the vial from liquid nitrogen tank and thaw cells quickly in a 37°C water-bath.
2. Just before the cells are completely thawed, decontaminate the outside of the vial with 70% ethanol and transfer the cells to a 15 ml centrifuge tube containing 9 ml of complete growth medium.
3. Pellet cells by centrifugation at 200 x g force for 5 min, and remove the medium.
4. Resuspend the cells in complete growth medium.
5. Transfer the cell suspension to a 10 cm dish with 10 ml of complete growth medium.

6. Grow the cells in incubator with 37°C, 5 %CO_2.
7. In the following day, replace the cells with fresh medium contains antibiotic.

Sub-culturing Protocol

1. Remove the culture medium from cells.
2. Wash cells with PBS (pH=7.4) to remove all traces of serum that contains trypsin inhibitor.
3. Add 2.0 ml of 0.05% (w/v) Trypsin- EDTA (GIBCO, Cat No. 25300) solution into 10 cm dish and observe the cells under an inverted microscope until cell layer is dispersed (usually within 3 to 5 minutes).
   Note: To avoid cells clumping, do not agitate the cells by hitting or shaking the dish while waiting for the cells detach. If cells are difficult to detach, please place the dish in 37°C incubator for ~2 min.
4. Add 6.0 to 8.0 ml of complete growth medium into dish and aspirate cells by gently pipetting.
5. Centrifuge the cells at 200 x g force for 5min, and remove the medium.
6. Resuspend the cells in culture medium and add the cells suspension to new culture dish.
7. Grow the cells in incubator with 37°C, 5 %CO_2.

Subcultivation Ratio: 1:3 to 1:8 weekly.
Medium Renewal: Every 2 to 3 days

V. REFERENCES

1. Hou C, *et al.* Subtype-specific signaling mechanisms of somatostatin receptors SSTR1 and SSTR2. *J Biol Chem.* 1994 Apr 8; 269(14):10357-62.
2. Schindler M, *et al.* Cellular localisation and co-expression of somatostatin receptor messenger RNAs in the human brain. *Brain Res Mol Brain Res.* 1995 Dec 28; 34(2):321-6.
3. Buscail L, *et al.* Stimulation of tyrosine phosphatase and inhibition of cell proliferation by somatostatin analogues: mediation by human somatostatin receptor subtypes SSTR1 and SSTR2. *Proc Natl Acad Sci U S A.* 1994 Mar 15; 91(6):2315-9.
4. Adams RL, *et al.* Inhibition of endothelial proliferation by the somatostatin analogue SOM230. *Clin Endocrinol (Oxf).* 2004 Oct; 61(4):431-6.

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