

**Human Recombinant SST2 Somatostatin Receptor Stable Cell Line****Cat. No. M00325****Version 06092014**

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**I. INTRODUCTION**

Catalog Number: M00325

Cell Line Name: CHO-K1/SST2/Gα15

Gene Synonyms: SSTR2

Expressed Gene: Genbank Accession Number NM\_001050; no expressed tags

Host Cell: CHO-K1/Gα15

Quantity: Two vials of frozen cells ( $3 \times 10^6$  per vial)

Stability: 16 passages

Application: Functional assay for SST2 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, 400 µg/ml Zeocin, 200 µg/ml Hygromycin B

Mycoplasma Status: Negative

Storage: Liquid nitrogen immediately upon delivery

**II. BACKGROUND**

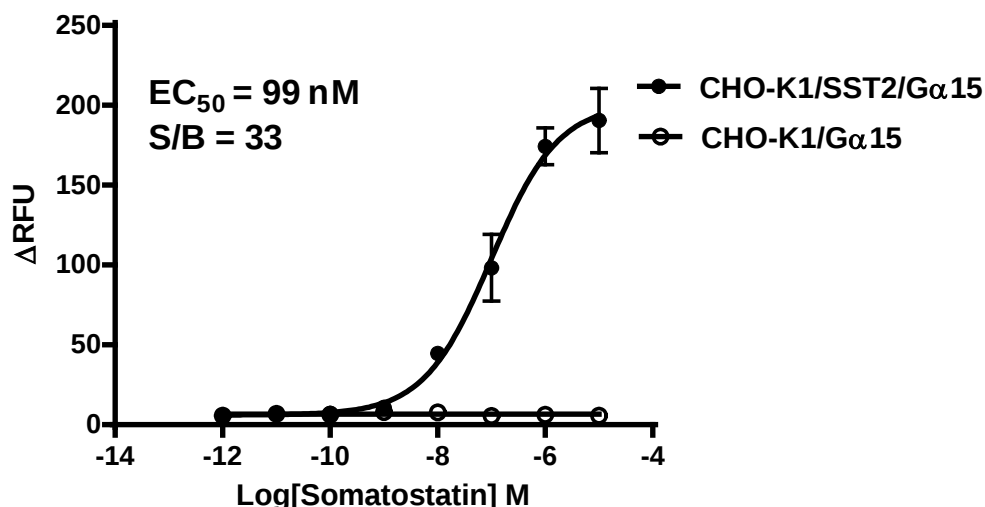
Somatostatin acts at many sites to inhibit the release of many hormones and other secretory proteins. The biologic effects of somatostatin are probably mediated by a family of G protein-coupled receptors that are expressed in a tissue-specific manner. SST2 is a member of the superfamily of receptors having seven transmembrane segments and is expressed in highest levels in cerebrum and kidney. Studies showed the involvement of the SST2 receptor in the inhibition of glucagon secretion.

§: 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.

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### III. REPRESENTATIVE DATA

Concentration-dependent stimulation of intracellular calcium mobilization by Somatostatin in CHO-K1/SST2/Gα15 and CHO-K1/Gα15 cells



**Figure 1.** Somatostatin-induced concentration-dependent stimulation of intracellular calcium mobilization in CHO-K1/SST2/Gα15 and CHO-K1/Gα15 cells. The cells were loaded with Calcium-4 prior to stimulation with an SST2 receptor agonist, Somatostatin. 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 (Mean ± SD, n = 2). The EC<sub>50</sub> of Somatostatin on SST2 co-expressing with Gα15 in CHO-K1 cells was 99 nM. The S/B of Somatostatin on SST2 co-expressing with Gα15 in CHO-K1 cells was 33.

#### Notes:

- EC<sub>50</sub> value is calculated with four parameter logistic equation:  

$$Y = \text{Bottom} + (\text{Top} - \text{Bottom}) / (1 + 10^{((\text{LogEC}_{50} - X) * \text{HillSlope}))}$$

X is the logarithm of concentration. Y is the response  
Y is RFU and starts at Bottom and goes to Top with a sigmoid shape.
- Signal to background Ratio (S/B) = Top/Bottom

### IV. THAWING AND SUBCULTURING

#### Thawing Protocol

- Remove the vial from liquid nitrogen tank and thaw cells quickly in a 37°C water-bath.
- 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.
- Pellet cells by centrifugation at 200 x g force for 5 min, and remove the medium.
- 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<sub>2</sub>.
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<sub>2</sub>.

Subcultivation Ratio: 1:3 to 1:8 weekly.

Medium Renewal: Every 2 to 3 days

## V. REFERENCES

1. Yamada Y, Stoffel M, Espinosa R 3rd, *et al.* (1993) Human somatostatin receptor genes: localization to human chromosomes 14, 17, and 22 and identification of simple tandem repeat polymorphisms. *Genomics* 15 (2): 449–52.
2. Zitzer, H; Hönck H H, Bächner D, *et al.* (1999) Somatostatin receptor interacting protein defines a novel family of multidomain proteins present in human and rodent brain. *J. Biol. Chem.* 274 (46): 32997–3001.

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