Human Recombinant H1 Histamine Receptor Stable Cell Line

Cat. No. M00131 **Version 05222014**

I. INTRODUCTION

Catalog Number: M00131
Cell Line Name: HEK293/H1
Gene Synonyms: HRH1, H1-R, hisH1
Expressed Gene: Genbank Accession Number NM_ 000861, no expressed tags
Host Cell: HEK293
Quantity: Two vials of frozen cells (3×10^6 per vial)
Stability: 16 passages
Application: Functional assay for H1 receptor
Freeze Medium: 45% culture medium, 45% FBS, 10% DMSO
Complete Growth Medium: DMEM, 10% FBS
Culture Medium: DMEM, 10% FBS, 200 μg/ml Zeocin
Mycoplasma Status: Negative
Storage: Liquid nitrogen immediately upon delivery

II. BACKGROUND

The H1 histamine receptor is expressed primarily in the lungs, vasculature, and brain. The H1 mediates the contraction of smooth muscles, neurotransmission in the central nervous system, the release of catecholamine from adrenal medulla, and increases in capillary permeability due to contraction of terminal venules. H1's role in inflammatory responses makes its antagonist suitable for treating allergies.

III. REPRESENTATIVE DATA

Concentration-dependent stimulation of intracellular calcium mobilization by Histamine in HEK293/H1 and HEK293 cells

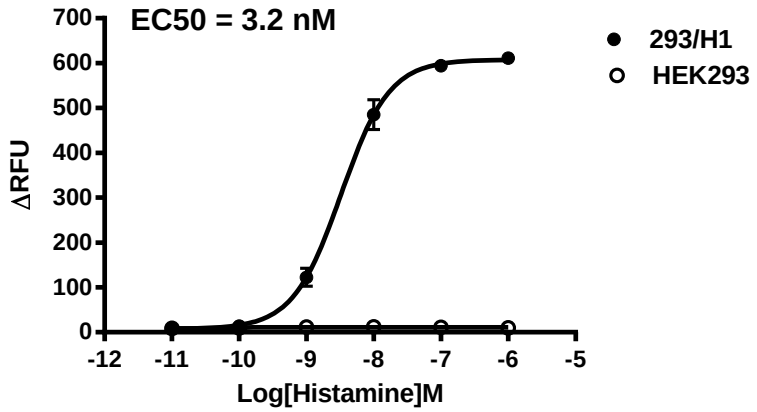

Figure Intracellular calcium response from HEK293 cells stably expressing human H1 histamine receptors and from untransfected control cells. Cells were loaded with Calcium-4 then stimulated with the indicated concentrations of histamine. Calcium responses were recorded on a FlexStation plate reader. Data represent the averge +/- standard deviation of triplicate determinations.

Note:

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.

V. REFERENCES

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GenScript USA Inc,
860 Centennial Ave.
Piscataway, NJ 08854
Toll-Free: 1-877-436-7274
Tel: 1-732-885-9188, Fax: 1-732-210-0262
Email: product@genscript.com
Web: http://www.genscript.com

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