Human Recombinant $G_{\alpha 15}$ Stable Cell Line
Cat. No. M00554 **Version 03232016**

I INTRODUCTION 1
II BACKGROUND 1
III REPRESENTATIVE DATA 2
IV THAWING AND SUBCULTURING 2
Limited Use License Agreement 4

I. INTRODUCTION

Catalog Number: M00554

Cell Line Name: HEK293/$G_{\alpha 15}$

Gene Synonyms: GNA15, GNA16

Official Full Name: Guanine nucleotide binding protein (G protein), alpha 15 (Gq class)

Expressed Gene: GenBank Accession Number NM_002068; no expressed tags

Host Cell: HEK293

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

Stability: 16 passages

Application: Functional assay for Gs and Gi/o-coupled GPCR receptors

Freeze Medium: 90% FBS, 10% DMSO

Complete Growth Medium: DMEM, 10% FBS

Culture Medium: DMEM, 10% FBS, 0.5 µg/ml puromycin

Mycoplasma Status: Negative

Storage: Liquid nitrogen immediately upon delivery

II. BACKGROUND

1) HEK293/$G_{\alpha 15}$

HEK293/$G_{\alpha 15}$ is a HEK293 cell line stably expressing the $G_{\alpha 15}$ alpha subunit protein which a Gq protein. It is used as a host cell for transfection expression of Gs and Gi/o -coupled receptors, the constitutively expressed $G_{\alpha 15}$ protein in the cells allows many transfected receptors which normally stimulate/inhibit the cAMP pathway, to couple to Gq signal transduction and mobilize intracellular calcium. The cell line carries the puromycin resistance gene and is resistant to puromycin

§: 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.

2) The sequence of $G_{\alpha15}$

ATGGCCCGCTCGCTGACCTGGCGCTGCTGCCCCTGGTGCCTGACGGAGGATGAGAAGGCCGCCGCCCGGG
TGGACCAGGAGATCAACAGGATCCTCTTGGAGCAGAAGAAGCAGGACCGCGGGGAGCTGAAGCTGCTGCT
TTTGGGCCCAGGCGAGAGCGGGAAGAGCACCTTCATCAAGCAGATGCGGATCATCCACGGCGCCGGCTAC
TCGGAGGAGGAGCGCAAGGGCTTCCGGCCCCTGGTCTACCAGAACATCTTCGTGTCCATGCGGGCCATGA
TCGAGGCCATGGAGCGGCTGCAGATTCCATTCAGCAGGCCCGAGAGCAAGCACCACGCTAGCCTGGTCAT
GAGCCAGGACCCCTATAAAGTGACCACGTTTGAGAAGCGCTACGCTGCGGCCATGCAGTGGCTGTGGAGG
GATGCCGGCATCCGGGCCTACTATGAGCGTCGGCGGGAATTCCACCTGCTCGATTCAGCCGTGTACTACC
TGTCCCACCTGGAGCGCATCACCGAGGAGGGCTACGTCCCCACAGCTCAGGACGTGCTCCGCAGCCGCAT
GCCCACCACTGGCATCAACGAGTACTGCTTCTCCGTGCAGAAAACCAACCTGCGGATCGTGGACGTCGGG
GGCCAGAAGTCAGAGCGTAAGAAATGGATCCATTGTTTCGAGAACGTGATCGCCCTCATCTACCTGGCCT
CACTGAGTGAATACGACCAGTGCCTGGAGGAGAACAACCAGGAGAACCGCATGAAGGAGAGCCTCGCATT
GTTTGGGACTATCCTGGAACTACCCTGGTTCAAAAGCACATCCGTCATCCTCTTTCTCAACAAAACCGAC
ATCCTGGAGGAGAAAATCCCCACCTCCCACCTGGCTACCTATTTCCCCAGTTTCCAGGGCCCTAAGCAGG
ATGCTGAGGCAGCCAAGAGGTTCATCCTGGACATGTACACGAGGATGTACACCGGGTGCGTGGACGGCCC
CGAGGGCAGCAAGAAGGGCGCACGATCCCGACGCCTCTTCAGCCACTACACATGTGCCACAGACACACAG
AACATCCGCAAGGTCTTCAAGGACGTGCGGGACTCGGTGCTCGCCCGCTACCTGGACGAGATCAACCTGC
TGTGA

III. REPRESENTATIVE DATA

Intracellular calcium mobilization of 10 µM VIP on transient-transfected HEK293/Gα15 cells with pcDNA3.1-VPAC2 plasmid

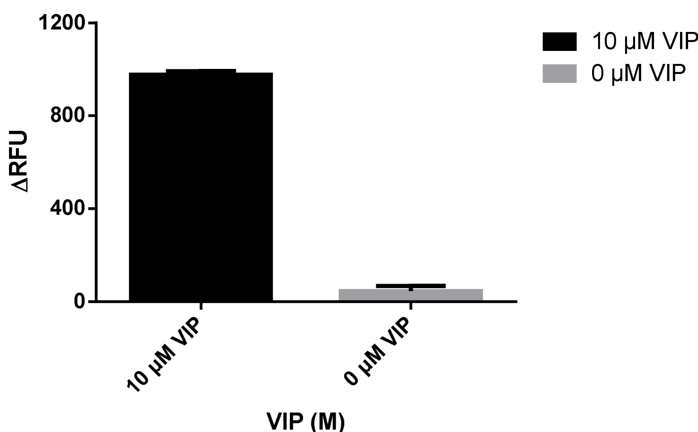

Figure 1. Intracellular calcium mobilization of 10 µM VIP on transient-transfected HEK293/$G_{\alpha15}$ cells with pcDNA3.1-VPAC2 plasmid.

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

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. Re-suspend 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

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

For Research Use Only.

Limited Use License Agreement

This is a legal agreement between you (Licensee) and GenScript USA Inc. governing use of GenScript's stable cell line products and protocols provided to licensee. By purchasing and using the stable cell line, the buyer agrees to comply with the following terms and conditions of this label license and recognizes and agrees to such restrictions:

1) The products are not transferable and will be used at the site where they were purchased. Transfer to another site owned by buyer will be permitted only upon written request by buyer followed by subsequent written approval by GenScript.
2) The purchaser cannot sell or otherwise transfer (a) this product (b) its components or (c) materials made using this product or its components to a third party.
3) The products sold by GenScript are for laboratory and animal research purposes only. The products are not to be used on humans, for consumption, or for any unlawful uses.

GenScript USA Inc. will not assert against the buyer a claim of infringement of patents owned or controlled by GenScript USA Inc. and claiming this product based upon the manufacture, use or sale of a clinical diagnostic, therapeutic and vaccine, or prophylactic product developed in research by the buyer in which this product or its components has been employed, provided that neither this product nor any of its components was used in the manufacture of such product. For information on the use of this product for other purposes, contact Marketing Department, GenScript USA Inc., 120 Centennial Avenue, Piscataway, New Jersey 08840, U.S.A. Phone: 1-732-885-9188. Fax: 1-732-210-0262. Email: marketing@genscript.com.