Human Recombinant Gqi5 Stable Cell Line

Cat. No. M00455 **Version 07102018**

I. Introduction

Catalog Number: M00455
Cell Line Name: CHO-K1/Gqi5
Host Cell: CHO-K1
Quantity: Two vials of frozen cells (>1×10^6 per vial)
Application: Functional assay for Gi/o-coupled GPCR receptors
Freeze Medium: 45% culture medium, 45% FBS, 10% DMSO
Complete Growth Medium: Ham's F12, 10% FBS
Culture Medium: Ham's F12, 10% FBS, 100 μg/ml Hygromycin B
Mycoplasma Status: Not detected*
Storage: Liquid nitrogen immediately upon delivery

II. Background

1. CHO-K1/Gqi5

CHO-K1/Gqi5 is a CHO-K1 cell line stably expressing the chimeric Gqi5 alpha subunit protein which a chimeric Gq protein instead of the last five carboxyl-terminal amino acids from Gi. It is used as a host cell for transfection expression of Gi/o-coupled receptors, the constitutively expressed Gqi5 protein in the cells allows many transfected receptors which normally inhibit the cAMP pathway, to couple to Gq signal transduction and mobilize intracellular calcium. The cell line carries the hygromycin B resistance gene and is resistant to hygromycin B.

2. The sequence of Gqi5

Atggcccgctcgctgacctggcgctgctgcccctggtgcctgacggaggatgagaaggccgccgcccgggtggaccaggagatcaa
caggatcctcttggagcagaagaagcaggaccgcggggagctgaagctgctgcttttgggcccaggcgagagcgggaagagcacc
ttcatcaagcagatgcggatcatccacggcgccggctactcggaggaggagcgcaagggcttccggcccctggtctaccagaacatct
tcgtgtccatgcgggccatgatcgaggccatggagcggctgcagattccattcagcaggcccgagagcaagcaccacgctagcctgg
tcatgagccaggacccctataaagtgaccacgtttgagaagcgctacgctgcggccatgcagtggctgtggagggatgccggcatccg
ggcctactatgagcgtcggcgggaattccacctgctcgattcagccgtgtactacctgtcccacctggagcgcatcaccgaggagggct
acgtccccacagctcaggacgtgctccgcagccgcatgcccaccactggcatcaacgagtactgcttctccgtgcagaaaaccaacct
gcggatcgtggacgtcgggggccagaagtcagagcgtaagaaatggatccattgtttcgagaacgtgatcgccctcatctacctggcct
cactgagtgaatacgaccagtgcctggaggagaacaaccaggagaaccgcatgaaggagagcctcgcattgtttgggactatcctgg
aactaccctggttcaaaagcacatccgtcatcctctttctcaacaaaaccgacatcctggaggagaaaatccccacctcccacctggcta
cctatttccccagtttccagggccctaagcaggatgctgaggcagccaagaggttcatcctggacatgtacacgaggatgtacaccggg
tgcgtggacggccccgagggcagcaagaagggcgcacgatcccgacgcctcttcagccactacacatgtgccacagacacacaga
acatccgcaaggtcttcaaggacgtgcgggactcggtgctcgcccgctacctggacgagTGTGGCCTCTTCTGA

** The mycoplasma test was performed with MycoAlert™ PLUS Mycoplasma Detection Kit of Lonza.*

GenScript USA, Inc.
860 Centennial Ave. Piscataway, NJ 08854 Tel: 1-877-436-7274; 1-732-885-9188

III. 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 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. Add antibiotic the following day.

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 0.25% (w/v) Trypsin- EDTA (GIBCO, Cat No. 25200-072) 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 for 5 min, 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: 1:4 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.