

Host Gqs5-Coupled CHO Cells (CHO/Gqs5)

Order Number: RD-HGQS5

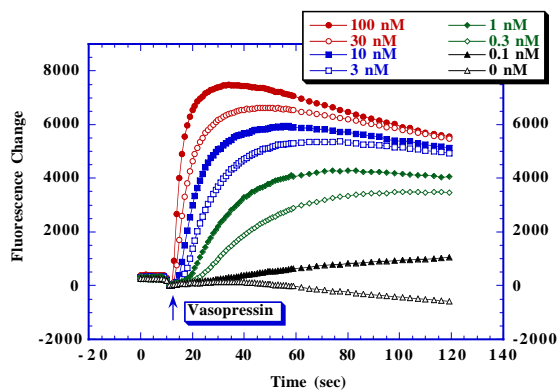
Cell Density and Storage: 2.4×10^6 cells frozen in 80 μ l of DMSO-free freezing medium. Cells must be stored in liquid nitrogen if not immediately processed upon receipt.

Note: A License from Molecular Devices Corporation is required prior to purchasing or using the cell line.

A. Description

CHO/Gqs5 is a CHO cell line stably expressing the chimeric Gqs5 alpha subunit protein. When used as a host cell for transfection expression of Gs-coupled receptors, the constitutively expressed Gqs5 protein in the cells allows some transfected receptors, which normally act through the cAMP pathway, to couple to Gq signal transduction and mobilize intracellular calcium. The cell line carries the hygromycin resistance gene and is resistant to hygromycin B.

Functional testing was carried out by transient transfection of the Gs-coupled V2 vasopressin receptor into the CHO/Gqs5 host cells and measuring the stimulation of intracellular calcium mobilization by vasopressin.



Coupling of V2 vasopressin receptor to intracellular calcium mobilization in CHO/Gqs5 cells. V2 vasopressin receptor cDNA was transiently transfected into CHO/Gqs5 cells using Lipofectamine. The transfected cells were loaded with Fluo-3 and vasopressin-mediated changes in intracellular calcium were measured by the FLIPR™ instrument (Molecular Devices Corporation, Sunnyvale, CA).

B. Thawing and Propagation of CHO/Gqs5 Cells

B.1. Culture Medium

Ham's F12 culture medium containing 10% fetal bovine serum, 100 unit/ml penicillin, 100 µg/ml streptomycin, 2 mM glutamine, and 200 µg/ml hygromycin B.

B.2. Thawing and Seeding

1. Remove cell vial from the liquid nitrogen container.
2. Place vial in a 37°C heating block and immediately add 80 µl of culture medium to the vial . Allow to thaw, mix gently, and equilibrate at 37°C for 5 min.
3. Dilute the thawed cells with 0.5 ml of culture medium and seed into a T-75 flask containing 25 ml of culture medium. Grow the cells in a CO₂ incubator at 37°C.

B.3. Subculture and Propagation

1. Split the cells 1:10 every 3 to 4 days, using trypsin-EDTA to harvest the cells.
2. It is strongly recommended that a frozen cell bank be established soon after thawing the cells and establishing the initial cultures.