DiscoverX

cAMP Hunter™ CHO-K1 EBI2 Gi Cell Line

Catalog Number: 95-0172C2

Contents:

Background

Lot Number:

See Vial

2 vials, 1 x 10⁶ cells per vial in 1 mL

cAMP Hunter™ Gi cell lines overexpress naturally Gi coupled, wildtype GPCRs and are designed to detect inhibition of intracellular cAMP production in response to agonist stimulation of the receptor. These cell lines are designed to be used in conjunction with the HitHunter® cAMP Assay Detection Kit.

Product Information

Target GPCR:	EBI2
Description:	G protein-coupled receptor 183
Receptor Family:	Class A Orphan
Coupling:	Gi/Go
Accession Number:	NM_004951.4
GPCR Species:	Human
Cell Type:	CHO-K1
Storage:	Short term (<24 h): Store at -80°C; Long term (>24 h): Store in vapor phase of liquid nitrogen.

Functional Performance

Cells were plated in a 384-well plate and incubated overnight at 37°C and 5% CO₂ to allow the cells to attach and grow. Cells were then stimulated with a control agonist + forskolin (Figure 1A) or with forskolin only (Figure 1B), using the assay conditions described below. Following stimulation, signal was detected using the HitHunter cAMP Assay Detection Kit according to the recommended protocol. Please refer to page 2 for recommended assay reagents, detection reagents, and control compounds.

Cell Number/Well:	10000	
Control Agonist:	7α,25-dihydroxycholesterol	
Agonist Incubation Time (minutes):	30	
Agonist Incubation Temperature (°C):	37	
Figure 1A. Agonist Dose Response		Figure 1B. Forskolin Dose Response
$ \begin{array}{c} 20000 \\ 15000 \\ 10000 \\ 5000 \\ 0 \\ 10^{-11} \\ 10^{-10} \\ 10^{-9} \\ 10^{-9} \\ 10^{-9} \\ 10^{-7} \\ 10^{-7} \\ 10^{-5} \\ 10^{-5} \\ 10^{-4} \\ 7\alpha, 25 \\ - dihydroxycholesterol [M] \\ + 10 \ \mu M \ Forskolin \end{array} $	Ē	25000 20000 15000- 10000- 5000- 0- 10 ⁻¹⁰ 10 ⁻⁸ 10 ⁻⁸ 10 ⁻⁷ 10 ⁻⁶ 10 ⁻⁵ 10 ⁻⁴ 10 ⁻³ 10 ⁻² Forskolin [M]
EC_{50} for Agonist Stimulation (nM):	634.2 EC₅₀ f	or Forskolin Stimulation (μM): 8694
Signal:Background at Agonist E _{max} : 3	3.7 Signa	al:Background at Forskolin E _{max} : 18.2
Forskolin Concentration (µM): 1	10	

Important! This assay requires an additional step: Please refer to Additional Protocol Information section.

1

DiscoverX

Passage Stability

This cell line has been confirmed to be stable through 10 passages with no significant drop in assay window or change in EC_{50} .

Mycoplasma Testing

This lot was tested and found to be free of mycoplasma contamination. Data available upon request.

Required Materials

The following additional materials are required but not provided:

Product Use*	Product Description	Catalog Number
Detection	HitHunter [®] cAMP Assay for Small Molecules	90-0075SM Series
	HitHunter cAMP Assay for Biologics	90-0075LM Series
Cell Culture	AssayComplete™ Cell Culture Kit-107	92-3107G
Cell Plating	AssayComplete™ Cell Plating 21 Reagent	93-0563R21A
Cell Detachment	AssayComplete™ Cell Detachment Reagent	92-0009
Cell Thawing	AssayComplete™ Thawing Reagent T2	92-4102TR
Cell Freezing	AssayComplete™ Freezing Reagent F2	92-5102FR

*Please inquire about our cell line-specific AssayComplete Starter Packs to get you started with your cell culture needs.

Required Antibiotics

Antibiotic Name	Concentration (µg/mL)	Catalog Number
AssayComplete™ Puromycin	Not Applicable	Not Applicable
AssayComplete™ Hygromycin B	Not Applicable	Not Applicable
AssayComplete™ G418	800	92-0030

Additional Ligand Information

Control Agonist: 7α ,25-dihydroxycholesterol **Vendor:** DiscoverX[®] (Catalog No. 92-1186)

Additional Protocol Information

It is recommended that the control compound be serially diluted in a base media (or buffer) + 1% FBS to ensure optimal assay performance.

For order placement or technical support, please call 1.866.448.4864 (North America) or +44.121.260.6142 (Europe) or e-mail info@discoverx.com. For additional information, please visit discoverx.com.

2

DiscoverX

Limited Use License Agreement

These products may be covered by issued US and/or foreign patents, patent application and subject to Limited Use Label License.

Please visit discoverx.com/license for a list of products that are governed by limited use label license terms and relevant patent and trademark information.