

PathHunter® EBI2 Total GPCR Internalization U2OS Cell Line

Catalog Number: 93-0910C3 **Lot Number:** See Vial
Contents: 2 vials, 1 x 10⁶ cells per vial in 1 mL

Background

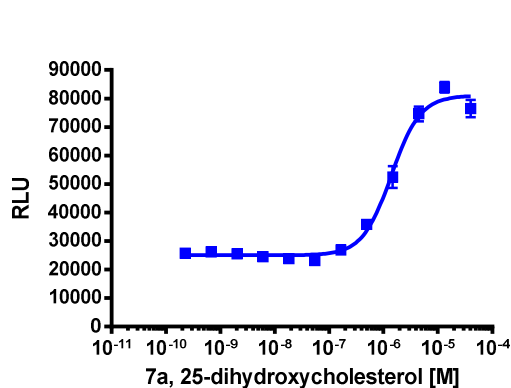
PathHunter Total GPCR Internalization cell lines are engineered to co-express the ProLink™ (PK) tagged GPCR, and an Enzyme Acceptor (EA) tag localized to the endosomes. Activation of the GPCR-PK induces internalization of the receptor in EA-tagged endosomes, forcing complementation of the two β-galactosidase enzyme fragments (EA and PK). The resulting functional enzyme hydrolyzes substrate to generate a chemiluminescent signal.

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: U2OS
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, using the assay conditions described below. Following stimulation, signal was detected using the PathHunter Detection Kit according to the recommended protocol. Please refer to page 2 for recommended assay reagents, detection reagents, and control compounds.



Cell Number/Well: 5000
Control Agonist: 7α,25-dihydroxycholesterol
Agonist Incubation Time (minutes): 180
Agonist Incubation Temperature (°C): RT
EC₅₀ for Agonist Stimulation (nM): 1400
Signal:Background at Agonist E_{max}: 3.0

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

Passage Stability

This cell line has been confirmed to be stable through 10 passages with no significant drop in assay window or change in EC₅₀.

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	PathHunter® Detection Kit	93-0001
Cell Culture	AssayComplete™ Cell Culture Kit-112	92-3112G
Cell Plating	AssayComplete™ Cell Plating 19 Reagent	93-0563R19A
Cell Detachment	AssayComplete™ Cell Detachment Reagent	92-0009
Cell Thawing	AssayComplete™ Thawing Reagent T3	92-4103TR
Cell Freezing	AssayComplete™ Freezing Reagent F3	92-5103FR

*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	N/A	N/A
AssayComplete™ Hygromycin B	250	92-0029
AssayComplete™ G418	500	92-0030

Additional Ligand Information

Control Agonist: 7α,25-dihydroxycholesterol

Vendor: DiscoverX® (Catalog No. 92-1186)

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.

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