

PathHunter® U2OS KCNH2(G601S) MEM-EA Pharmacotraficking Assay

Catalog Number: 93-1064C3 **Lot Number:** See Vial

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

Background

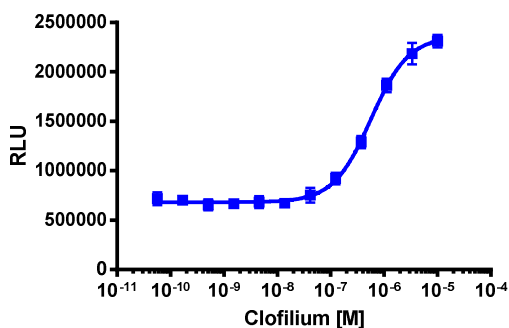
PathHunter Pharmacotraficking cell lines are engineered to co-express a ProLink™ (PK)-tagged transmembrane protein retained in the ER (due to protein misfolding), and an Enzyme Acceptor (EA) tag localized to the cell membrane. Binding of a chemical pharmacochaperone to the misfolded, PK-tagged protein stabilizes the protein in a conformation that enables its trafficking through the Golgi, then onward to the cell membrane. Localization of the protein to the EA-tagged cell membrane forces 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 Protein: KCNH2(G601S)
Description: Potassium voltage-gated channel, subfamily H (eag-related), member 2
Accession Number: NM_000238
Target Species: Human
ProLink™ Tag: PK1
Readout: Membrane - EA
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 for at least 4 hours at 37°C and 5% CO₂ to allow the cells to attach. Cells were then stimulated with a control compound, 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 Compound: Clofilium
Compound Incubation Time (minutes): Overnight
Compound Incubation Temperature (°C): 37
EC₅₀ for Compound Stimulation (nM): 564
Signal:Background at Agonist E_{max}: 3.5

Please Note: The PathHunter KCNH2(G601S) Pharmacotraficking Cell Line grows unusually slowly, particularly during the first few passages. 1:2 - 1:3 split ratios are recommended for passaging cells.

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® Flash Detection Kit	93-0247
Cell Culture	AssayComplete™ Cell Culture Kit-103	92-3103G
Cell Plating	AssayComplete™ Cell Plating 5 Reagent	93-0563R5A
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	Not Applicable	Not Applicable
AssayComplete™ Hygromycin B	250	92-0029
AssayComplete™ G418	500	92-0030

Additional Ligand Information

Control Compound: Clofilium

Vendor: DiscoverX® (Catalog No. 92-1302)

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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