

# PathHunter® CHO-K1 EDG7 $\beta$ -Arrestin Cell Line

**Catalog Number:** 93-0636C2

**Lot Number:** See Vial

**Contents:** 1 x 10<sup>6</sup> cells per vial in 1 mL

## Background

PathHunter  $\beta$ -Arrestin GPCR cell lines are engineered to co-express the ProLink™ (PK) tagged GPCR and the Enzyme Acceptor (EA) tagged  $\beta$ -Arrestin. Activation of the GPCR-PK induces  $\beta$ -Arrestin-EA recruitment, forcing complementation of the two enzyme fragments. The resulting functional enzyme hydrolyzes substrate to generate a chemiluminescent signal. PathHunter cell lines expressing Gq-coupled receptors can also be used to detect calcium mobilization.

## Product Information

**Target GPCR:** EDG7

**Description:** Lysophosphatidic acid receptor 3

**Receptor Family:** Lysophospholipid (LPA)

**$\beta$ -Arrestin Isoform:**  $\beta$ -Arrestin-2

**Accession Number:** NM\_012152.2

**ProLink™ Tag:** PK1

**Coupling:** Gq

**Cell Type:** CHO-K1

**GPCR Species:** Human

**Storage:** Short term (<24 h): Store at -80°C; Long term (>24 h): Store in vapor phase of liquid nitrogen.

## Functional Performance

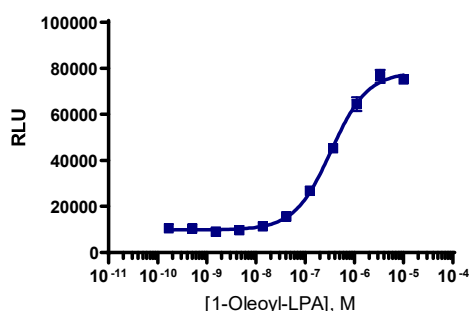
Cells were plated in a 384-well plate and incubated overnight at 37°C and 5% CO<sub>2</sub> to allow the cells to attach and grow. Cells were then stimulated with a control agonist using the assay conditions described below. Following stimulation, signal from Arrestin recruitment was detected using the PathHunter Detection Kit according to the recommended protocol. Calcium mobilization was detected using the Calcium No Wash<sup>PLUS</sup> detection kit. Please refer to page 2 for recommended assay reagents, detection reagents, and control compounds.

### PathHunter Arrestin Assay

**Cell Number/Well:** 5000

**Agonist Incubation Time (minutes):** 180

**Agonist Incubation Temp. (°C):** RT



**EC<sub>50</sub> for Arrestin Recruitment (nM):** 335

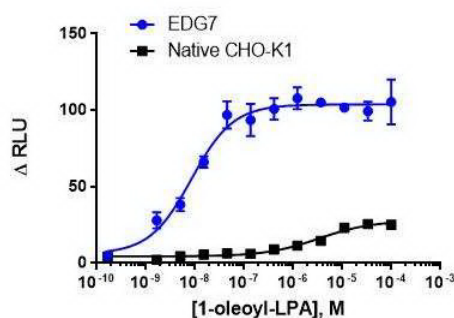
**Signal:Background at agonist E<sub>max</sub>:** 8.2

### Ca<sup>2+</sup> Mobilization Assay

**Cell Number/Well:** 10000

**Signal Read Time (@ 2 sec intervals):** 2 minutes

**Signal Read Temperature (°C):** RT



**EC<sub>50</sub> for Calcium Mobilization (nM):** 8.5

**Note:** This cell line was developed and quality control tested via the PathHunter Arrestin Assay only. Calcium mobilization assay was run independently from the PathHunter Arrestin Assay. Calcium data is background subtracted and represented as  $\Delta$ RFU (Relative Fluorescence Units).

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

### Passage Stability

This cell line has been confirmed to be stable through a minimum of 10 passages with no significant drop in assay window or change in EC<sub>50</sub>. Passage stability testing was conducted using the PathHunter Arrestin Assay only.

### 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        |
| Ca <sup>2+</sup> Detection | Calcium No Wash <sup>PLUS</sup>        | 90-0091        |
| Cell Culture               | AssayComplete™ Cell Culture Kit-108    | 92-3108G       |
| Cell Plating               | AssayComplete™ Cell Plating 13 Reagent | 93-0563R13A    |
| 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 | 300                   | 92-0029        |
| AssayComplete™ G418         | 800                   | 92-0030        |

### Additional Ligand Information

**Control Compound:** 1-Oleoyl-LPA

**Vendor:** Eurofins DiscoverX® (Catalog No. 92-1041)

### Additional Protocol Information

If running a Calcium flux assay with these cells, it is important that the buffer (e.g. HBSS/20mM HEPES) used as the diluent for the agonist dose-response dilution curve (or EC80 challenge) contains 0.1% BSA.

**Ordering:** +1.510.979.1415 option 4 or e-mail [CustomerServiceDRX@eurofins.com](mailto:CustomerServiceDRX@eurofins.com)

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