

## PathHunter® CHO-K1 ADORA3 $\beta$ -Arrestin Cell Line

**Catalog Number:** 93-0586C2 **Lot Number:** See Vial

**Contents:** 2 vials,  $1 \times 10^6$  cells per vial in 1 mL

### Background

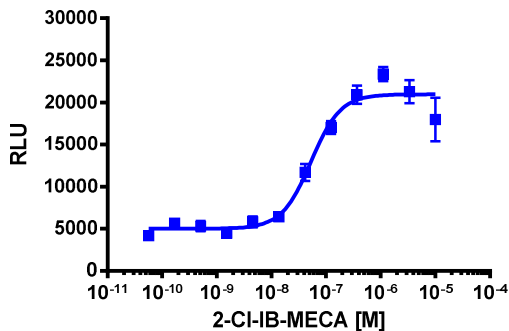
PathHunter  $\beta$ -Arrestin GPCR cell lines are engineered to co-express the ProLink™ (PK) tagged GPCR and the Enzyme Acceptor (EA) tagged  $\beta$ -Arrestin. Activation of the GPCR-PK induces  $\beta$ -Arrestin-EA recruitment, forcing complementation of the two  $\beta$ -galactosidase enzyme fragments (EA and PK). The resulting functional enzyme hydrolyzes substrate to generate a chemiluminescent signal.

### Product Information

**Target GPCR:** ADORA3  
**Description:** Adenosine receptor A3  
**Receptor Family:** Adenosine  
**Coupling:** Gi/Go  
**Accession Number:** NM\_000677.3  
**GPCR Species:** Human  
 **$\beta$ -Arrestin Isoform:**  $\beta$ -Arrestin-2  
**ProLink™ Tag:** ARMS2-PK2  
**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% 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 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:** 2-CI-IB-MECA  
**Agonist Incubation Time (minutes):** 90  
**Agonist Incubation Temperature (°C):** 37  
**EC<sub>50</sub> for Agonist Stimulation (nM):** 53.1  
**Signal:Background at Agonist E<sub>max</sub>:** 5  
**Control Antagonist:** VUF 5574  
**IC<sub>50</sub> of Antagonist (nM):** 14.8

### 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<sub>50</sub>.

### 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-107    | 92-3107G       |
| Cell Plating    | AssayComplete™ Cell Plating 2 Reagent  | 93-0563R2A     |
| 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:** 2-CI-IB-MECA

**Vendor:** DiscoverX® (Catalog No. 92-1045)

### Additional ProLink™ Tag Description

PK2 is a slight variant of PK1 and has been shown to enhance EFC. ARMS (Arrestin Recruitment Modulating Sequence) is an 18-21 amino acid spacer between the GPCR and the PK tag and has been shown to enhance β-Arrestin recruitment.

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