

## PathHunter<sup>®</sup> CHO-K1 rCHRM4 $\beta$ -Arrestin Cell Line

**Catalog Number:** 93-0870C2 **Lot Number:** See Vial

**Contents:** 2 vials, 1 x 10<sup>6</sup> cells per vial in 1 mL

### Background

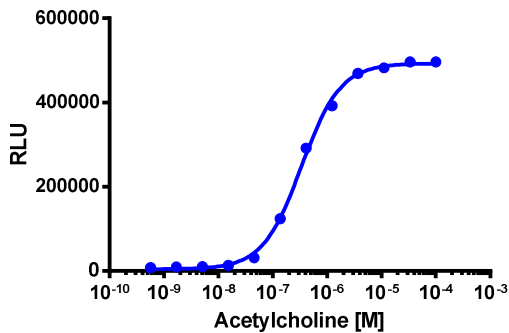
PathHunter  $\beta$ -Arrestin GPCR cell lines are engineered to co-express the ProLink<sup>™</sup> (PK) tagged GPCR and the Enzyme Acceptor (EA) tagged  $\beta$ -Arrestin. Activation of the GPCR-PK induces  $\beta$ -Arrestin-EA recruitment, forcing complementation of the two  $\beta$ -galactosidase enzyme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

### Product Information

**Target GPCR:** rCHRM4  
**Description:** Cholinergic receptor, muscarinic 4  
**Receptor Family:** Acetylcholine  
**Coupling:** Gs & Gi/Go  
**Accession Number:** NM\_031547  
**GPCR Species:** Rat  
 **$\beta$ -Arrestin Isoform:**  $\beta$ -Arrestin-2  
**ProLink<sup>™</sup> 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:** Acetylcholine  
**Agonist Incubation Time (minutes):** 90  
**Agonist Incubation Temperature (°C):** 37  
**EC<sub>50</sub> for Agonist Stimulation (nM):** 339  
**Signal:Background at Agonist E<sub>max</sub>:** 62.8

### 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 <sup>®</sup> 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:** Acetylcholine

**Vendor:** DiscoverX<sup>®</sup> (Catalog No. 92-1125)

### 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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