

PathHunter® U2OS PRLR-JAK1 Functional Assay

Catalog Number: 93-0686C3 **Lot Number:** See Vial

Contents: 2 vials, 4 x 10⁶ cells per vial in 1 mL

Background

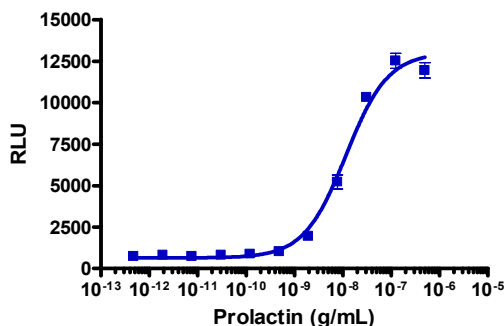
PathHunter Cytosolic Tyrosine Kinase (CTK) Functional cell lines are engineered to co-express a ProLink™ (PK) tagged Cytokine receptor, an untagged CTK, and an Enzyme Acceptor (EA) tagged SH2 domain. Activation of the Receptor-PK induces phosphorylation of the receptor by the CTK. This leads to SH2-EA recruitment, 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

| | | | |
|------------------------------|---|-------------------------|----------------|
| Cytokine Receptor: | PRLR | Target CTK: | JAK1 |
| Accession #: | NM_000949 | CTK Accession #: | NM_002227 |
| Receptor Description: | Prolactin receptor | CTK Description: | Janus kinase 1 |
| Receptor Species: | Human | CTK Species: | Human |
| SH2 Domain: | PLCG2 | | |
| 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: | 10000 |
| Control Agonist: | Prolactin |
| Ligand Incubation Time (minutes): | 180 |
| Ligand Incubation Temperature (°C): | RT |
| EC₅₀ for SH2 recruitment (ng/mL): | 11.6 |
| Signal:Background at agonist E_{max}: | 17.2 |

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-103 | 92-3103G |
| Cell Plating | AssayComplete™ Cell Plating 0 Reagent | 93-0563R0A |
| 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 | 0.25 | 92-0028 |
| AssayComplete™ Hygromycin B | 250 | 92-0029 |
| AssayComplete™ G418 | 500 | 92-0030 |

Additional Ligand Information

Control Agonist: Prolactin

Vendor: DiscoverX® (Catalog No. 92-1144)

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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