

PathHunter® U2OS FcγRIIb Clustering Cell Line

Catalog Number: 93-1133C3

Lot Number: See Vial

Contents: 1 x 10⁶ cells per vial in 1 mL

Background

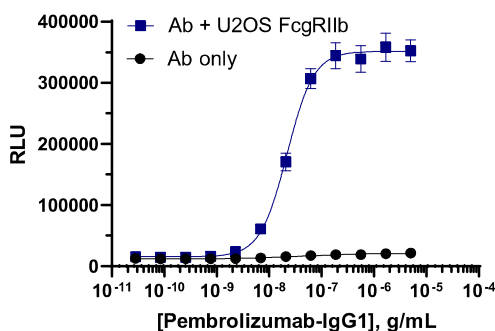
Checkpoint Clustering Cell Lines are companion products to the Checkpoint Signaling Assays that have been engineered to exogenously express a specific Fcγ receptor. When co-cultured with an appropriate Checkpoint Signaling Cell Line in the presence of antigen-specific agonist antibodies, the Fcγ receptor binds to the Fc portion of the antibody, presenting the antigen binding end to the checkpoint receptor on the Checkpoint Signaling Cell Line. This results in receptor clustering and robust receptor activation, leading to downstream signaling events, which are detected by β-galactosidase complementation. The resulting functional enzyme hydrolyzes substrate to generate a chemiluminescent signal.

Product Information

Target: FcγRIIb
Accession #: NM_004001.4
Description: Low affinity immunoglobulin gamma Fc region receptor II-b
Species: human
Cell Type: U2OS (Adherent)
Storage: Short term (<24 h): Store at -80°C; Long term (>24 h): Store in vapor phase of liquid nitrogen.

Functional Performance

Fcγ receptor cells were seeded in a 96-well plate and then pre-incubated with a control antibody, followed by co-incubation with the signaling assay cell line 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.



Clustering Cell Number/Well: 10000

Control Antibody: Anti-hPD1-Pem-hlgG1

Antibody Pre-Incubation Time (minutes): 60

Antibody Pre-Incubation Temperature (°C): 37

Signaling Assay: PathHunter® Jurkat PD-1 (SHP1) Signaling Cell Line

Signaling Assay Cell Number/Well: 20000

Target:Clustering Cell Ratio: 2:1

Signaling Cell Inc Time (minutes): 120

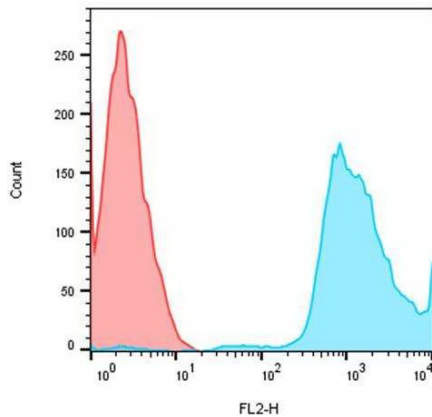
Signaling Cell Temperature (°C): RT

EC₅₀ for Antibody (ng/mL): 21.9

Signal:Background at Agonist E_{max}: 24.9

Detection Incubation Time (hrs.) 1

Cell Line Fcg Receptor Expression



Fcg receptor clustering cells were immunostained for receptor expression and analyzed by flow cytometry. Red, native U2OS; blue, U2OS Fcg receptor clustering cell line.?

Passage Stability

This cell line has been confirmed to be stable through a minimum of 10 passages with no significant drop in expression levels.

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 |
|-----------------|--|----------------|
| 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 | 0.5 | 92-0028 |
| AssayComplete™ Hygromycin B | Not Applicable | Not Applicable |
| AssayComplete™ G418 | Not Applicable | Not Applicable |

To be used with: PathHunter® Jurkat PD-1 (SHP1) Signaling Cell Line (93-1104C19)

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