

PathHunter® U2OS mCD80 Ligand Cell Line

Catalog Number: 93-1162C3

Lot Number: See Vial

Contents: 1.2 x 10⁶ cells per vial in 1 mL

Background

Checkpoint Ligand Cell Lines are companion products to the Checkpoint Signaling Assays, which have been engineered to exogenously express a cell surface ligand for a specific checkpoint receptor. When co-cultured with its cognate receptor-expressing cells, the ligand expressed on the Ligand Cell Line will engage with the checkpoint receptor on the Checkpoint Signaling Cell Line. This receptor-ligand engagement leads to receptor activation and downstream signaling events, which are detected by forcing complementation of the two β-galactosidase enzyme fragments: EA-tagged SH2 domain and PK-tagged checkpoint receptor. The resulting functional enzyme hydrolyzes substrate to generate a chemiluminescent signal.

Product Information

Target Ligand: mCD80

Accession #: NM_001359898

Description: T-lymphocyte activation antigen CD80

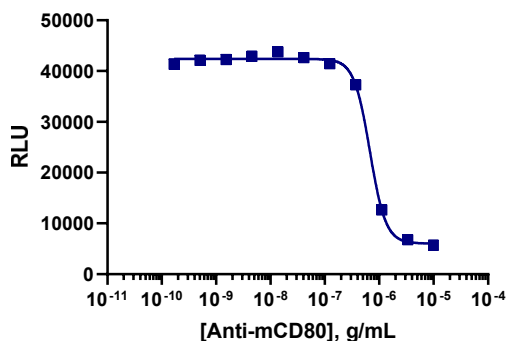
Species: Mouse

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

Signaling assay cells were seeded in a 96-well plate and then pre-incubated with a control antibody, followed by co-incubation with the ligand 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.



Signaling Assay:	PathHunter® Jurkat mCTLA4 Signaling Assay
Signaling Assay Cell Number/Well:	20000
Control Antibody:	
Antibody Pre-Incubation Time (minutes):	60
Antibody Pre-Incubation Temperature (°C):	37
IC₅₀ for Antibody (ng/mL):	673.7
Signal:Background at agonist E_{max}:	7.3
Ligand Cell Number/Well:	30000
Receptor:Ligand Cell Ratio:	1:1.5
Ligand Incubation Time (minutes):	300
Ligand Incubation Temperature (°C):	37
Detection Incubation Time (hrs.)	1

Note: For this assay, U2OS mCD80 Ligand cells were seeded first and pre-incubated for 1 hour at 37°C with Anti-mCD80 Antibody, followed by co-culture with the Jurkat mCTLA4 Signaling cells. Overnight (16 hour) incubation with detection reagent is recommended but the assay can also be measured after a one hour incubation.

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
Detection	PathHunter [®] Bioassay Detection Kit	93-0933
Cell Culture	AssayComplete™ Cell Culture Kit-103	92-3103G
Cell Plating	Not included with Ligand cell lines	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	Not Applicable	Not Applicable
AssayComplete™ G418	Not Applicable	Not Applicable

To be used with: PathHunter[®] Jurkat mCTLA4 Signaling Assay (93-1161C19)

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