

InCELL Hunter™ U2OS BCL2L1-BAX Protein Binding Cell Line

Catalog Number: 96-0009C3 **Lot Number:** See Vial

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

Background

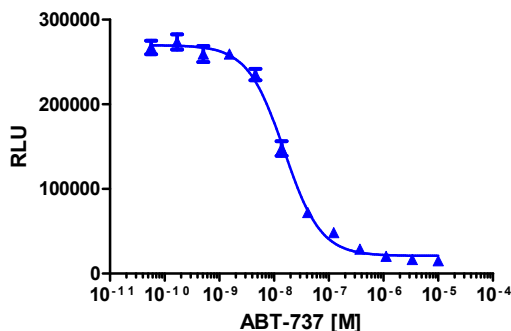
This InCELL Hunter assay measures intracellular compound-target engagement by detecting degradation of a partner protein that is ordinarily stabilized by its natural interaction with the target protein. The cell lines are engineered to co-express an untagged target protein, and an Enzyme Donor (ED) tagged partner protein. Either protein may be full length or truncated. Addition of inhibitors disrupting the interaction of the two proteins results in degradation of the ED-tagged protein. Addition of exogenous Enzyme Acceptor (EA), and buffer, lyses the cell and forces complementation of the ED and EA enzyme fragments. This results in the formation of a functional enzyme that hydrolyzes substrate to generate a chemiluminescent signal.

Product Information

Target Protein 1:	BCL2L1	Target Protein 2:	BAX
Amino Acid Range:	1 - 233	Amino Acid Range:	52 - 72
Accession #:	NM_138578.1	Accession #:	NM_138761
Description:	BCL2-like 1	Description:	BCL2-associated X protein
Target Tag 1:	None	Target Tag 2:	ePL
Target Tag 1 located at (terminus):	N/A	Target Tag 2 located at (terminus):	N-Terminus
Target Species:	Human		
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 compound, using the assay conditions described below. Following treatment, signal was detected using the InCELL Hunter Detection Kit according to the recommended protocol using the conditions specified below. Please refer to page 2 for recommended assay reagents, detection reagents, and control compounds.



Cell Number/Well:	20000
Cell Seeding Time (Hours):	24
Control Compound:	ABT-737
Ligand Incubation Time (minutes):	180
Ligand Incubation Temperature (°C):	37
IC₅₀ of Inhibitor (nM):	15.2
Signal:Background at compound E_{max}:	18.0
1:4 EA Dilution Required?	No
Detection Incubation Time (hours):	1

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	InCELL Detection Kit	96-0079
Cell Culture	AssayComplete™ Cell Culture Kit-103	92-3103G
Cell Plating	AssayComplete™ Cell Plating 16 Reagent	93-0563R16A
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	500	92-0030

Additional Ligand Information

Control Compound: ABT-737

Vendor: DiscoverX® (Catalog No. 92-1151)

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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