

SPRINTer™ HCT-116 CDKN1A (p21) Protein Turnover Biosensor Cell Line

Catalog Number: 91-1006C047

Lot Number: See Vial

Contents: 2 x 10⁶ cells per vial in 1 mL

Background

SPRINTer Protein Turnover Biosensor cell lines are engineered to introduce the EFC Enzyme Donor (ED) into the endogenous locus of the desired target gene. Expression of the target gene from its native promoter results in production of an ED-tagged target protein. Treatment of the engineered Biosensor cells with therapeutics that promote turnover of the target protein produces a decrease in EFC signal. 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, allowing quantitation of drug-induced changes in ED-target protein levels.

Product Information

Cell Type: HCT-116

Species: Human

Target Protein: CDKN1A (p21)

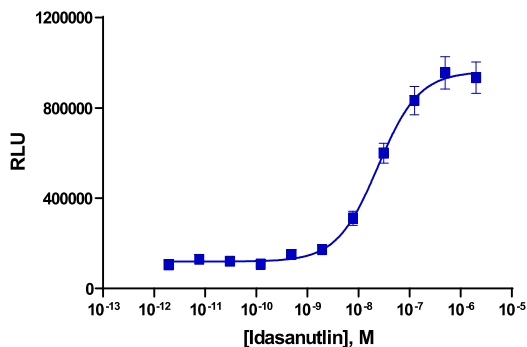
Description: cyclin dependent kinase inhibitor 1A

Target Tag: ePL

Tag Location: C-terminus

Functional Performance

Cells were plated in a 384-well plate and incubated 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 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

Cell Seeding Time (hours): 0

Control Agonist: Idasanutlin

Ligand Incubation Time (minutes): Overnight

Ligand Incubation Temperature (°C): 37

EC₅₀ for compound stimulation (nM): 24

Signal:Background at agonist E_{max}: 8.9

Note: For best assay performance:

- Trypsin or Cell Detachment Reagent should be removed by centrifugation when sub-culturing and before plating cells in the assay
- **2X10⁶** cells should be seeded into a 10 cm dish **two** days prior to lifting cells for plating in the assay

Passage Stability

This cell line has been confirmed to be stable through a minimum of 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 [®] ProLabel [®] /ProLink [™] Detection Kit	93-0812S
Cell Culture	AssayComplete [™] Cell Culture Kit-102	92-3102G
Cell Plating	AssayComplete [™] Cell Plating 0 Reagent	93-0563R0A
Cell Detachment	AssayComplete [™] Cell Detachment Reagent	92-0009
Cell Thawing	AssayComplete [™] Thawing Reagent T8	92-4108TR
Cell Freezing	AssayComplete [™] Freezing Reagent F6	92-5106FR

*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	Not Applicable	Not Applicable
AssayComplete [™] G418	Not Applicable	Not Applicable

Additional Ligand Information

Control Agonist: Idasanutlin

Vendor: Tocris (Catalog No. 6904)

Ordering: +1.510.979.1415 option 4 or e-mail CustomerServiceDRX@eurofins.com

Technical support: +1.510.979.1415 option 5 or e-mail DRX_SupportUS@eurofinsUS.com

General product information: www.discoverx.com

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