

SPRINTer eXpress BRAF(V600E) Protein Turnover Biosensor Assay (A375)

Catalog Number: 91-1008E057

Lot Number: See Vial

Contents: 1.2 x 10⁶ cells per vial in 0.1 mL

Background

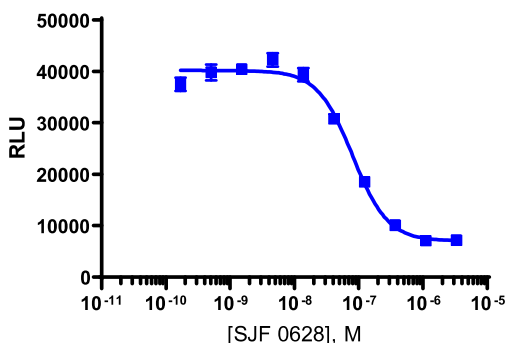
SPRINTer Protein Turnover Biosensor cells are engineered to introduce the EFC Enzyme Donor (ED) into the endogenous locus of the desired target gene. Expression of the target gene from its native promoter results in production of an ED-tagged target protein. Treatment of the engineered Biosensor cells 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. These cells have been modified to prevent long term propagation and expansion using a proprietary compound that has no apparent effect on assay performance.

Product Information

Cell Type: A375
Species: Human
Target Protein: BRAF(V600E)
Description: B-Raf proto-oncogene, serine/threonine kinase
Target Tag: ePL
Tag Location: C-Terminus

Functional Performance

Cells were plated in a 96-well plate and incubated at 37°C and 5% CO₂. Cells were then stimulated with a control compound, using the assay conditions described below. Following stimulation, signal was detected using the PathHunter Detection Reagents provided in the kit according to the recommended protocol. For a detailed protocol, please refer to the user manual.



Cell Number/Well: 10000
Cell Seeding Time (hours): 0
Control Agonist: SJF 0628
Ligand Incubation Time (minutes): 1080
Ligand Incubation Temperature (°C): 37
EC₅₀ for compound stimulation (nM): 80.3
Signal:Background at agonist E_{max}: 4.8

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