

ClariCELL™ ABL1 [T315I] Kinase Assay Service

Description

The ClariCELL™ ABL1 [T315I] Kinase Assay quantifies autophosphorylation of human full-length ABL1 [T315I] in human cells. The assay is useful to determine potencies of small-molecule inhibitors against the specified kinase in the context of a cellular environment. Compound testing services are available utilizing the assay.

Overview

Human Embryonic Kidney (HEK 293) cells transiently expressing sequence verified human full-length ABL1 [T315I] are exposed to test compound or control, then lysed to release cellular proteins. ABL1 [T315I] is captured onto an assay plate, and the extent of autophosphorylation is quantified by ELISA using an antibody specific for the phosphorylation event. Cells expressing kinase deficient ABL1 (K213M) are also utilized as controls to calculate the % inhibition of test compounds.

Assay Validation

ABL1 [T315I] Expression in Cells

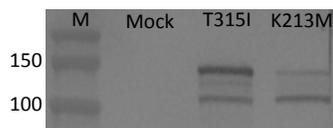


Figure 1: ABL1 [T315I] or kinase dead (K213M) ABL1 was expressed transiently in 293 cells. Following cell lysis, an IP Western was performed with appropriate antibodies to capture and detect total ABL1 protein.

ABL1 [T315I] Autophosphorylation in Cells

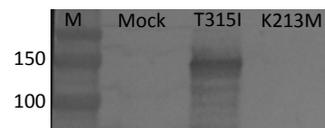


Figure 2: ABL1 [T315I] or kinase dead (K213M) ABL1 was expressed transiently in 293 cells. Following cell lysis, an IP Western was performed with appropriate antibodies to capture and detect phospho-ABL1 protein.

Quantification of Phosphorylation

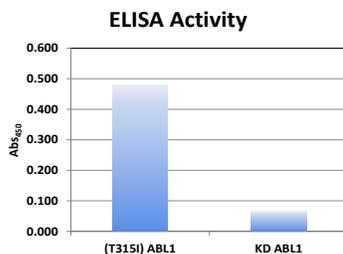


Figure 3: ABL1 [T315I] or kinase dead ABL1 (KD ABL1) was expressed transiently in 293 cells. Following cell lysis, an ELISA was performed to quantify the extent of auto-phosphorylation of ABL1 [T315I].

Reference Inhibitor Data

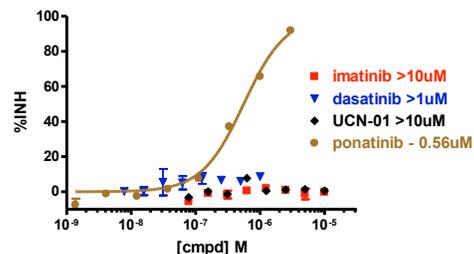


Figure 4: An autophosphorylation assay was performed in the presence of three ABL1 inhibitors (imatinib, dasatinib and ponatinib), and UCN-01, a compound that is not expected to inhibit ABL1 [T315I]. % inhibition data were plotted to determine EC₅₀s.