



# ClariCELL™ PIM-3/BAD Kinase Assay Service

## 4-Step Assay Validation

### PIM-3 Expression in Cells

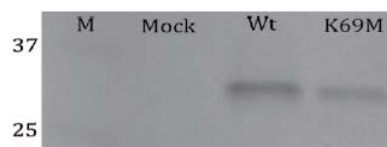


Figure 1: Wild type (Wt) or kinase dead (K69M) PIM-3 was expressed with BAD transiently in 293 cells. Following cell lysis, a Western was performed with appropriate antibodies to detect total PIM-3 protein.

### BAD Phosphorylation in Cells

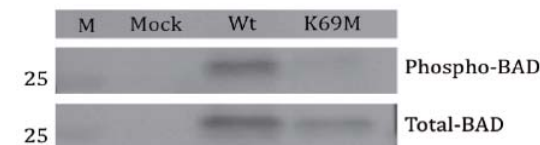


Figure 2: Wild type (Wt) or kinase dead (K69M) PIM-3 was expressed with BAD transiently in 293 cells. An IP Western was performed with appropriate antibodies to capture and detect phospho-BAD and total BAD protein.

### Description



The **ClariCELL™ PIM-3/BAD Kinase Assay** quantifies Pim-3 dependent phosphorylation of human full-length physiological substrate (BAD) 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 PIM-3 and BAD are exposed to test compound or control, then lysed to release cellular proteins. BAD is captured onto an assay plate, and the extent of phosphorylation is quantified by ELISA using an antibody specific for the phosphorylation event. Cells expressing kinase deficient PIM-3 [K69M] are also utilized as controls to calculate the % inhibition of test compounds.

### Quantification of Phosphorylation

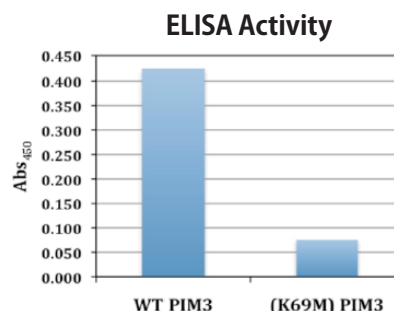


Figure 3: Wild type (WT) or kinase dead (K69M) PIM-3 was expressed with BAD transiently in 293 cells. Following cell lysis, an ELISA was performed to quantify the extent of phosphorylation of BAD.

### Reference Inhibitor Data

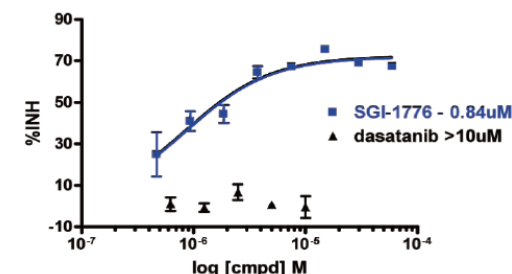


Figure 4: A phosphorylation assay was performed in the presence of SGI-1776, a PIM-3 inhibitor, and dasatanib, a compound that is not expected to inhibit PIM-3. % inhibition data were plotted to determine EC<sub>50</sub>s.

サービスの詳細、最新情報はHPをご覧ください

**カルナバイオサイエンス株式会社**

〒650-0047 神戸市中央区港島南町1-5-5 BMA3F

TEL: 078-302-7091 (営業部直通) / FAX: 078-302-7086

E-mail: [info@carnabio.com](mailto:info@carnabio.com)