NanoBRETTM Target Engagement Assay for PKN1

Protocol

HEK293 cells were transfected with PKN1-NanoLuc® Fusion Vector and seeded into the wells of 96-well plates. The cells were incubated with test compound for 2 hours following the addition of the NanoBRETTM tracer reagent. The BRET signal was measured on a luminometer after dispensing the NanoBRETTM Nano-Glo® Substrate and Extracellular NanoLuc® Inhibitor into the wells. IC50 values were calculated by fitting the data to the following eaquation.

BRET ratio = Min + (Max - Min)
$$\frac{1}{(1 + (IC50/[Compound])^{Slope})}$$

