

AlphaLISA™ SureFire® Ultra™

Human and Mouse p-Raf-1 (Ser259) Detection Kit

Product number: ALSU-PRAF-B500, ALSU-PRAF-B10K,
ALSU-PRAF-B50K, ALSU-PRAF-B-HV



Kit specificity:

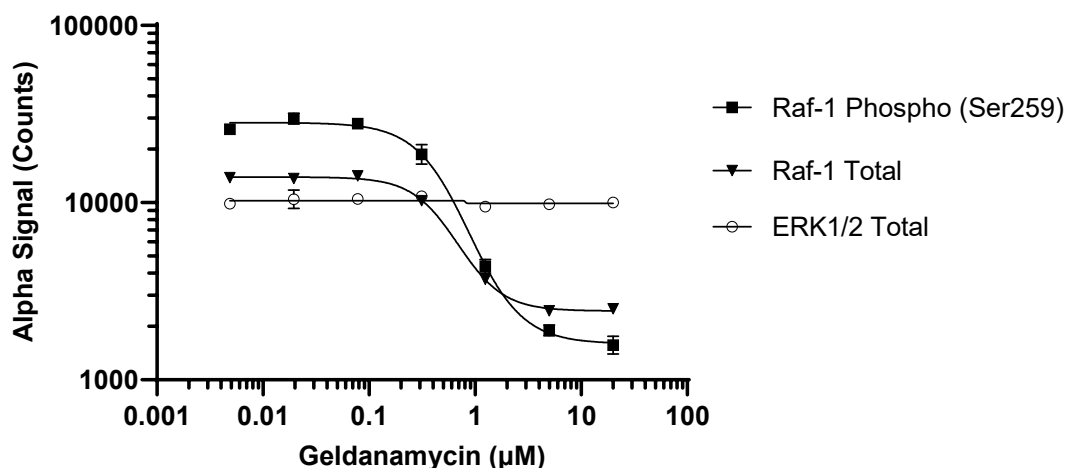
This assay kit contains antibodies which recognize the phospho- Ser259 epitope and a distal epitope on Raf-1. The protein detected by this kit corresponds to UniProt ID P04049. Raf-1 is also known as RAF proto-oncogene serine/threonine-protein kinase or Proto-oncogene c-RAF. These antibodies recognize Raf-1 of human and mouse origin. Other species should be tested on a case-by-case basis.

Control lysate information:

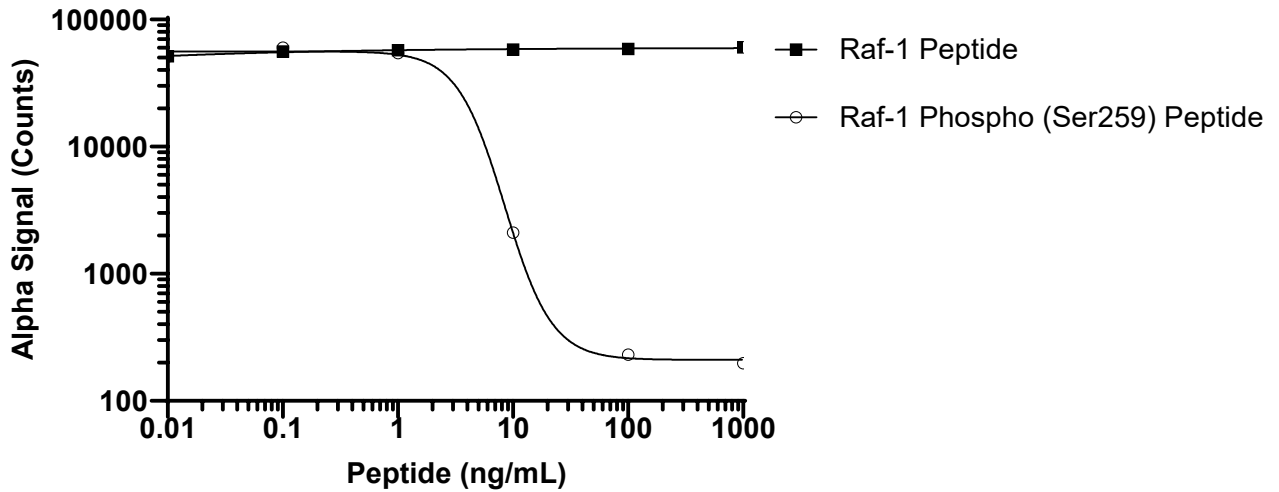
Positive Control Lysate: Prepared from HeLa cells, cultured to confluence in T175 flasks in 10 % FBS containing medium. Cells were treated with 200 ng/mL PMA for 10 minutes then lysed with 4 mL of Lysis Buffer.

Representative data:

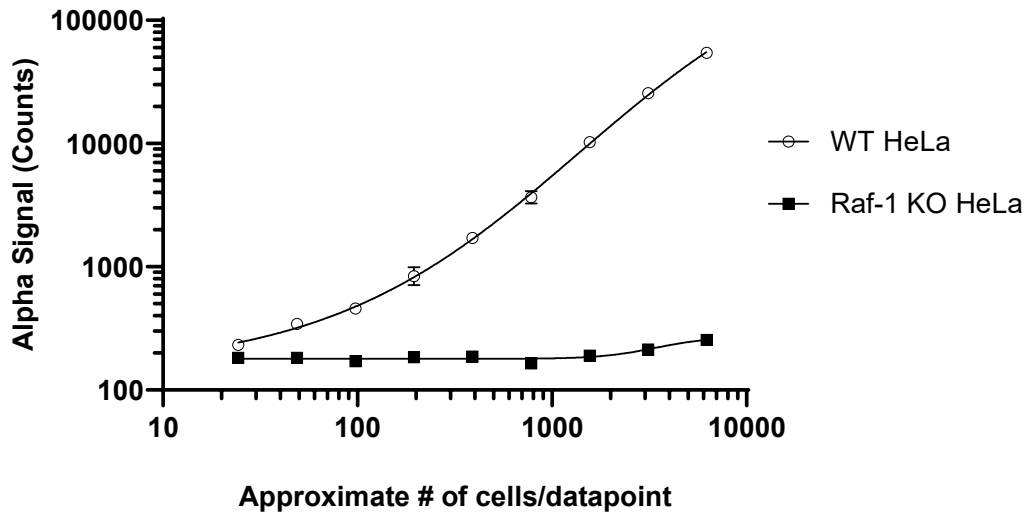
Data obtained by a 2-plate, 2-incubation protocol. HeLa cells were seeded at 40K cells/well in a 96 well plate and incubated overnight. Cells were then treated with Geldanamycin at the indicated concentrations for 24 hours in media containing 1% FBS. Cells were lysed with Lysis Buffer and assayed for various targets using respective SureFire Ultra kits. Equivalent to approximately 4,000 cells/datapoint.



Data obtained by a 1-plate, 2-incubation protocol. Phospho (Ser259) and Non-Phospho Raf-1 peptides were titrated into a fixed concentration of Raf-1 positive control lysate. Lysates were then assayed for Phospho Raf-1 (Ser259) using the *SureFire Ultra* kit. Data corresponds to approximately 8,000 cells/datapoint.



HeLa Raf-1 wild type (WT) and HeLa Raf-1 knockout (KO, Abcam ab264978) cell lines were cultured to confluence in T25 flasks. Cells were lysed with Lysis Buffer and assayed for Phospho (Ser259) Raf-1 using the *SureFire Ultra* assay kit. Approximate number of cells/datapoint is indicated.



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