AlphaLISA™ *SureFire® Ultra™*

Human and Mouse IRS1 Total Detection Kit

Product number: ALSU-TIRS1-A500, ALSU-TIRS1-A10K, ALSU-TIRS1-A50K, ALSU-TIRS1-A-HV

Kit specificity:

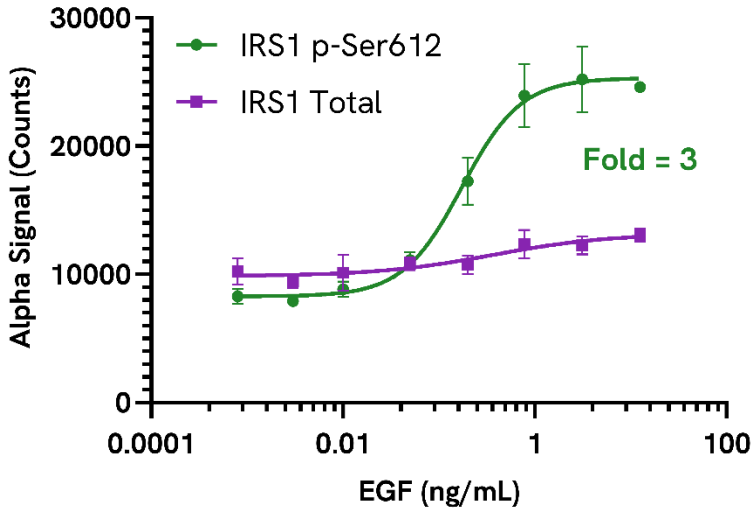
This assay kit contains antibodies which recognize distinct epitopes on IRS1. The protein detected by this kit corresponds to UniProt ID P35568. IRS1 is also known as Insulin receptor substrate 1. These antibodies recognize IRS1 of human and mouse origin. Other species should be tested on a case-by-case basis.

Control lysate information:

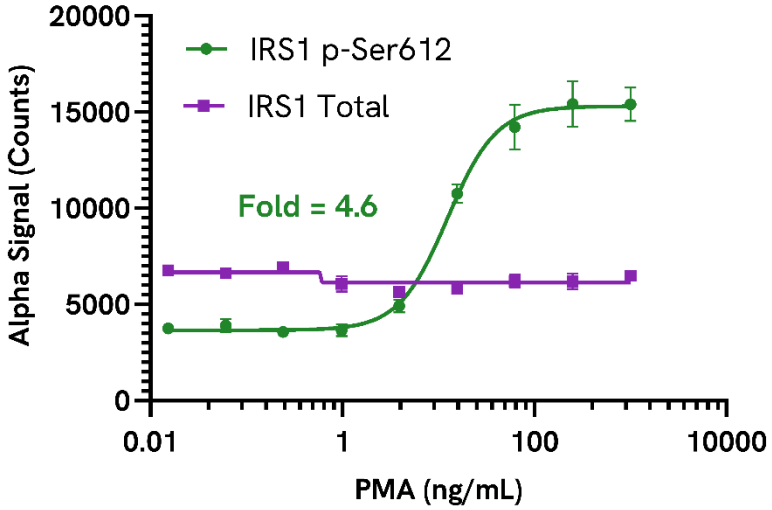
Positive Control Lysate: Prepared from MCF7 cells, cultured to confluence in T175 flasks in 10% FBS containing medium. Cells were lysed with 4 mL of Lysis Buffer.

Representative data:

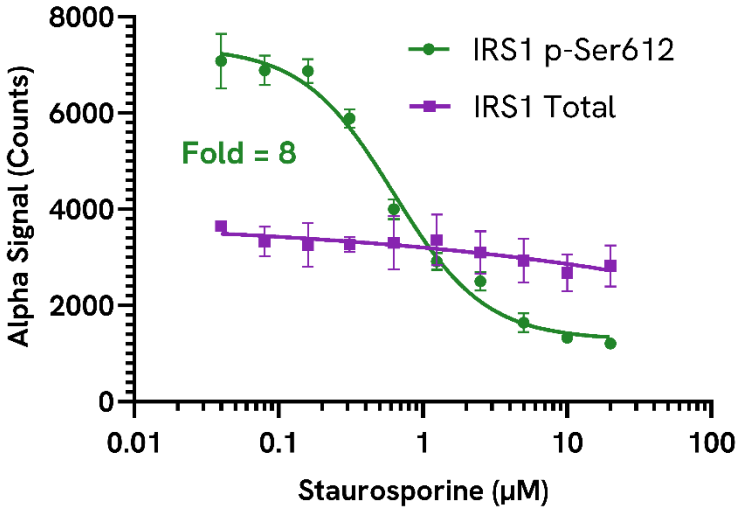
Data obtained with a 2-plate, 2-incubation protocol. A431 cells were seeded at 40K cells/well in a 96-well plate and incubated overnight. Cells were starved for 2 hours and treated with EGF at the indicated concentrations for 30 minutes. Cells were lysed with Lysis Buffer and assayed separately for Phospho (Ser612) and Total IRS1 using respective *SureFire Ultra* kits. Equivalent to approximately 4,000 cells/datapoint.



Data obtained with a 2-plate, 2-incubation protocol. HeLa cells were seeded at 40K cells/well in a 96-well plate and incubated overnight. Cells were starved for 24 hours and then treated with PMA at the indicated concentrations for 30 minutes. Cells were lysed with Lysis Buffer and assayed separately for Phospho (Ser612) and Total IRS1 using respective *SureFire Ultra* kits. Equivalent to approximately 4,000 cells/datapoint.



Data obtained with a 2-plate, 2-incubation protocol. A431 cells were seeded at 40K cells/well in a 96-well plate and incubated overnight. Cells were treated with Staurosporine at the indicated concentrations for 1 hour. Cells were lysed with Lysis Buffer and assayed separately for Phospho (Ser612) and Total IRS1 using respective *SureFire Ultra* kits. Equivalent to approximately 4,000 cells/datapoint.



Data obtained with a 2-plate, 2-incubation protocol. Adherent cell lines were seeded at 40K/well in a 96-well plate and incubated overnight. Suspension cell lines were seeded at 400K/well in HBSS + 0.1% BSA in a 96-well plate. Adherent and suspension cell lines were then lysed with Lysis Buffer and evaluated for Total IRS1 using the *SureFire Ultra* kit. Equivalent to approximately 4,000 cells/datapoint for adherent cell lines and 40,000 cells/datapoint for suspension cell lines.