AlphaLISA® *SureFire® Ultra™*

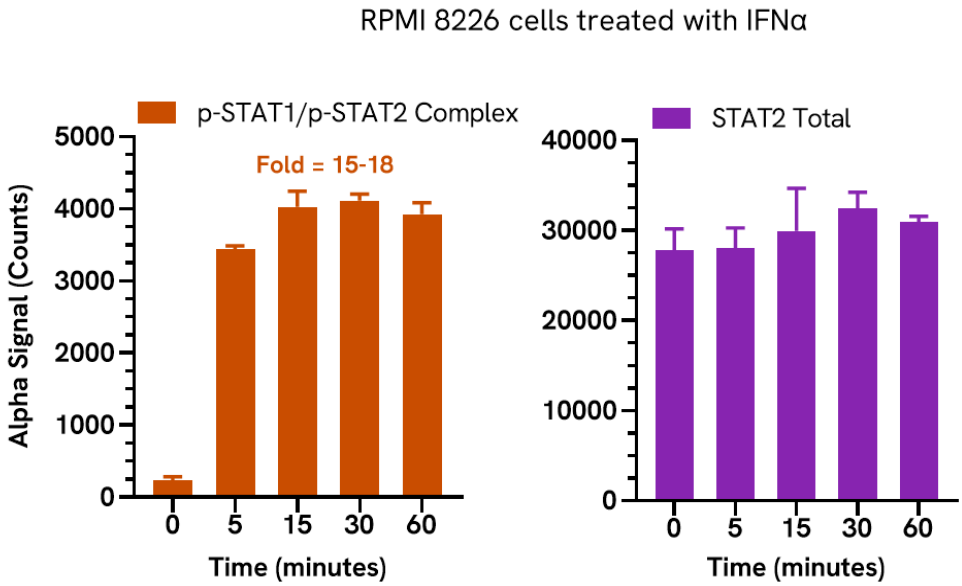
Human p-STAT1 (Tyr701) / p-STAT2 (Tyr690) Complex Detection Kit

Product number: ALSU-CST1ST2-A500, ALSU-CST1ST2-A10K, ALSU-CST1ST2-A50K, ALSU-CST1ST2-A-HV

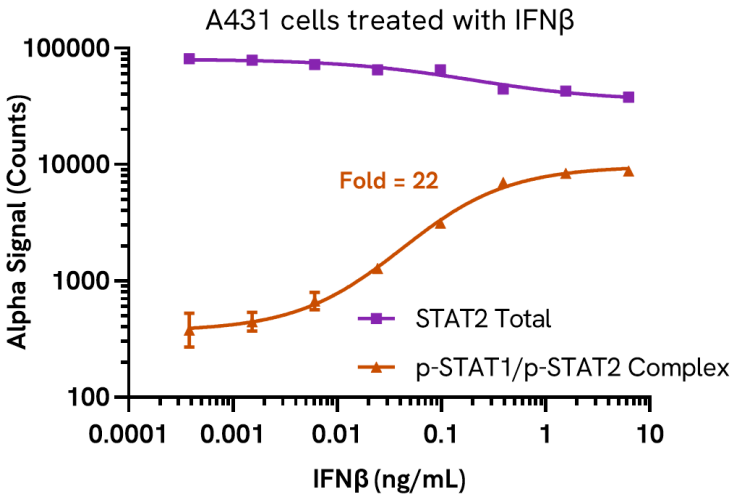
Kit specificity:
This assay kit contains antibodies which recognize the phospho-Tyr701 and phospho-Tyr690 epitopes on STAT1 and STAT2, respectively. The proteins detected by this kit correspond to UniProt ID P42224 (STAT1) and P27661 (STAT2). STAT1 is also known as signal transducer and activator of transcription 1-α/β and STAT2 is also known as signal transducer and activator of transcription 2. These antibodies recognize p-STAT1 and p-STAT2 of human origin. Other species should be tested on a case-by-case basis.

Control lysate information:
Positive Control Lysate: Prepared from A431 cells, cultured to confluence in T175 flasks in 10% FBS containing medium, then treated with 10 ng/mL recombinant human IFNβ for 30 minutes and lysed with 4 mL of Lysis Buffer.

Representative data:
Data obtained with a 2-plate, 2-incubation protocol. RPMI 8226 cells were harvested, seeded in a 96-well plate at 200K cells/well and treated with 10 ng/mL IFNα at the indicated timepoints. Cells were washed, lysed with Lysis Buffer and assayed separately for Phospho (Tyr701) STAT1/Phospho (Tyr690) STAT2 Complex and Total STAT2 using respective *SureFire Ultra* kits. Equivalent to approximately 20,000 cells/datapoint.



Data obtained with a 2-plate, 2-incubation protocol. A431 cells were seeded at 60K cells/well in a 96-well plate and incubated overnight. Cells were treated with IFNα at the indicated concentrations for 30 minutes. Cells were lysed with Lysis Buffer and assayed separately for Phospho (Tyr701) STAT1/Phospho (Tyr690) STAT2 Complex and Total STAT2 using respective *SureFire Ultra* kits. Equivalent to approximately 6,000 cells/datapoint.



Data obtained with a 2-plate, 2-incubation protocol. THP-1 cells were seeded at 200K cells/well in a 96-well plate with HBSS containing 10 ng/mL IFNβ for 15 minutes. Cells were then treated with the JAK inhibitor, Ruxolitinib at the indicated concentrations for 1 hour. Cells were lysed with Lysis Buffer and assayed separately for Phospho (Tyr701) STAT1/Phospho (Tyr690) STAT2 Complex and Total STAT2 using respective *SureFire Ultra* kits. Equivalent to approximately 20,000 cells/datapoint.