AlphaLISA™ *SureFire® Ultra™*

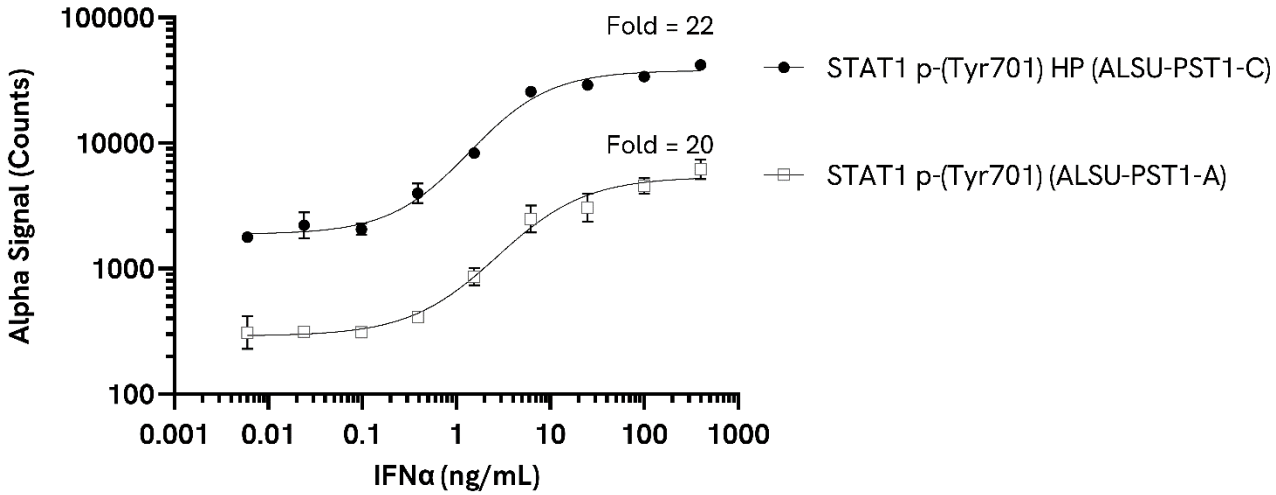
Human and Mouse p-STAT1 (Tyr701) High Performance Detection Kit

Product number: ALSU-PST1-C500, ALSU-PST1-C10K, ALSU-PST1-C50K, ALSU-PST1-C-HV

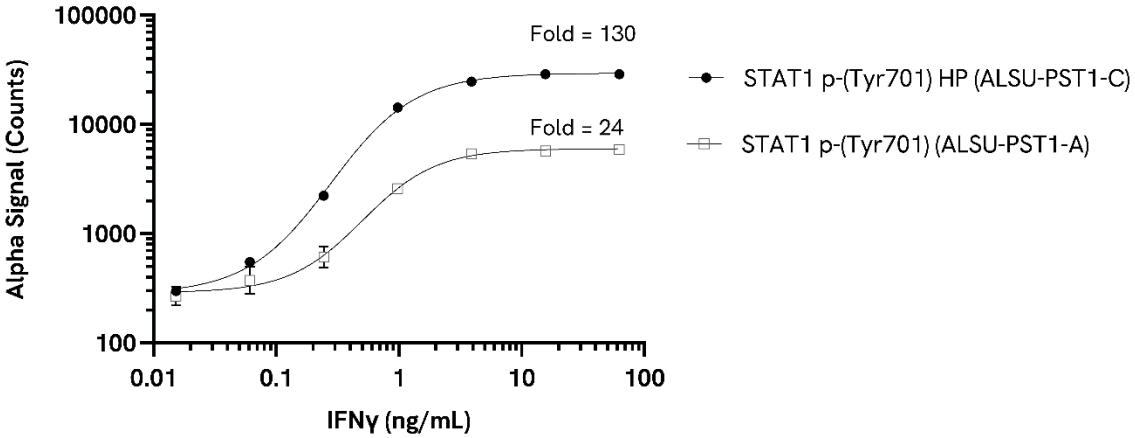
Kit specificity:
This assay kit contains antibodies which recognize the phospho-Tyr701 epitope and a distal epitope on STAT1. The protein detected by this kit corresponds to UniProt ID P42224. STAT1 is also known as signal transducer and activator of transcription 1-α/β. These antibodies recognize STAT1 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 0.6 M Sorbitol for 20 minutes followed by 200 ng/mL human IFNγ for a further 20 minutes and then lysed with 6 mL of Lysis Buffer.

Representative data:
Data obtained using a 2-plate, 2-incubation protocol. PBMCs were isolated from healthy donors and cultured for 6 days in complete DMEM containing 20 ng/mL M-CSF to differentiate them into macrophages. Macrophages were seeded at 30K cells/well in a 96-well plate and incubated overnight. Cells were treated with IFNα at the indicated concentrations for 10 minutes. Cells were lysed with Lysis Buffer and assayed for STAT1 Phospho (Tyr701) using *SureFire Ultra* kits ALSU-PST1-A and ALSU-PST1-C. Equivalent to approximately 2,000 (ALSU-PST1-A) or 400 (ALSU-PST1-C) cells/datapoint.



Data obtained using a 2-plate, 2-incubation protocol. RAW 264.7 cells were seeded at 40K cells/well in a 96-well plate and incubated overnight. The cells were starved in medium containing 1% FBS for 2 hours, then treated with mouse IFNγ at the indicated concentrations for 20 minutes. Cells were lysed with Lysis Buffer and assayed for STAT1 Phospho (Tyr701) using *SureFire Ultra* kits ALSU-PST1-A and ALSU-PST1-C. Equivalent to approximately 4,000 cells/datapoint.



Data obtained using a 2-plate, 2-incubation protocol. HEL92.1.7 cells were seeded at 400K cells/well in a 96-well plate and starved for 2 hours in HBSS + 0.1% BSA. Cells were treated with IFNα at the indicated concentrations for 10 minutes. Cells were lysed with Lysis Buffer and assayed separately for Phospho (Tyr701) HP and Total STAT1 using respective *SureFire Ultra* assay kits. Equivalent to approximately 4,000 cells/datapoint for Phospho (Tyr701) HP and 40,000 cells/datapoint for Total STAT1.