

AlphaLISA™ SureFire® Biotin Free

# Human IRF5 Aggregate Detection Kit

**Product number:** ASBF-AIRF5-A500, ASBF-AIRF5-A10K,  
ASBF-AIRF5-A50K, ASBF-AIRF5-A-HV



## Kit specificity:

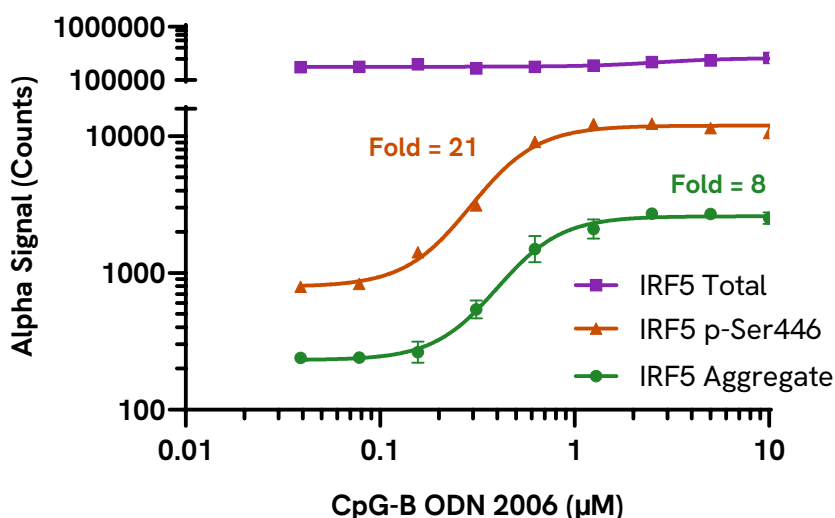
This assay kit contains antibodies which recognize aggregated IRF5. The protein detected by this kit corresponds to UniProt ID Q13568. IRF5 is also known as Interferon regulatory factor 5. These antibodies recognize IRF5 of human origin. Other species should be tested on a case-by-case basis.

## Control lysate information:

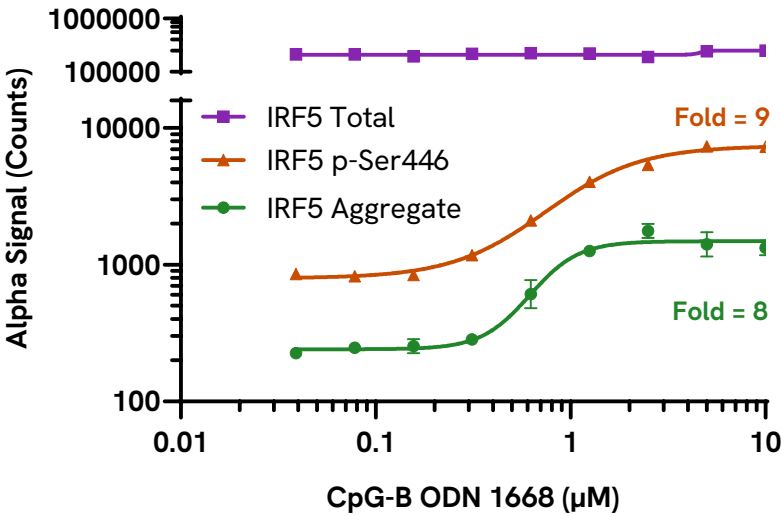
Positive Control Lysate: Prepared from THP-1 cells seeded at  $0.5 \times 10^6$  cells/mL and incubated overnight in 10% FBS containing medium. Cells were harvested, adjusted to  $5 \times 10^6$  cells/mL and treated with 100 nM Calyculin A for 3 hours. Following treatment, cells were washed with HBSS + 0.1% BSA and lysed with Lysis Buffer at a final concentration of  $10 \times 10^6$  cells/mL.

## Representative data:

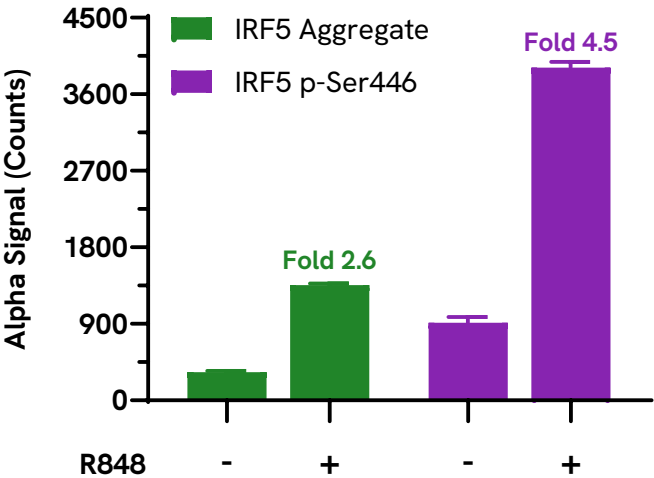
Data obtained with a 2-plate, 2-incubation protocol. RPMI 8226 cells were seeded at 200K/well in a 96-well plate. Cells were then treated with CpG-B ODN 2006 at the indicated concentrations for 3 hours. Cells were spun down at 1200 RPM for 5 minutes, supernatant was removed and cells were lysed with Lysis Buffer and assayed separately for Aggregate, Phospho (Ser446) and Total IRF5 using respective SureFire Biotin Free kits. Equivalent to approximately 40,000 cells/datapoint.



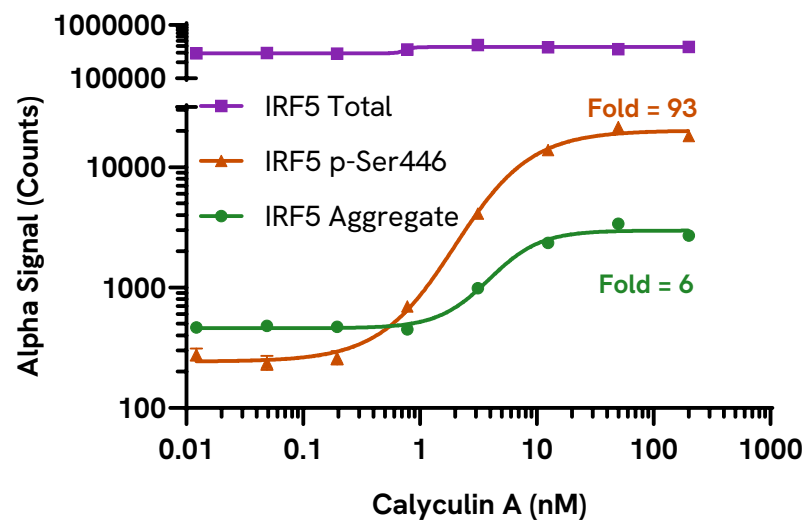
Data obtained with a 2-plate, 2-incubation protocol. RPMI 8226 cells were seeded at 200K/well in a 96-well plate. Cells were then treated with CpG-B ODN 1668 at the indicated concentrations for 3 hours. Cells were spun down at 1200 RPM for 5 minutes, supernatant was removed and cells were lysed with Lysis Buffer and assayed separately for Aggregate, Phospho (Ser446) and Total IRF5 using respective *SureFire* Biotin Free kits. Equivalent to approximately 40,000 cells/datapoint.



Data obtained with a 2-plate, 2-incubation protocol. RPMI 8226 cells were seeded at 400K/well in a 96 well plate. Cells were then treated with 2.5 μg/mL R848 (InvivoGen) for 4 hours. Cells were spun down at 1200 RPM for 5 minutes, supernatant was removed and cells were lysed with Lysis Buffer and assayed separately for Phospho (Ser446) and Aggregate IRF5 using respective *SureFire* Biotin Free kits. Equivalent to approximately 80,000 cells/datapoint.



Data obtained with a 2-plate, 2-incubation protocol. THP-1 cells were seeded at 200K/well in a 96-well plate. Cells were then treated with Calyculin A at the indicated concentrations for 3 hours. Cells were spun down at 1200 RPM for 5 minutes, supernatant was removed and cells were lysed with Lysis Buffer and assayed separately for Aggregate, Phospho (Ser446) and Total IRF5 using respective *SureFire* Biotin Free kits. Equivalent to approximately 40,000 cells/datapoint.



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