

AlphaLISA® SureFire® Ultra™

# Human p-CD3 Zeta (Tyr142) Detection Kit

**Product number:** ALSU-PCD3-B500, ALSU-PCD3-B10K,  
ALSU-PCD3-B50K, ALSU-PCD3-B-HV



**Kit specificity:**

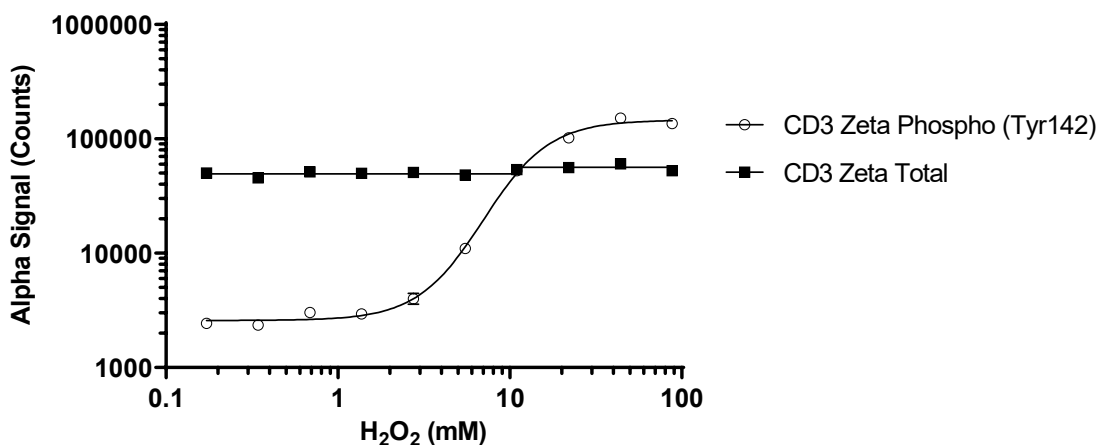
This assay kit contains antibodies which recognize the phospho-Tyr142 epitope and a distal epitope on CD3 Zeta. The protein detected by this kit corresponds to UniProt ID P20963. CD3 Zeta is also known as CD247. These antibodies recognize CD3 Zeta of human origin. Other species should be tested on a case-by-case basis.

**Control lysate information:**

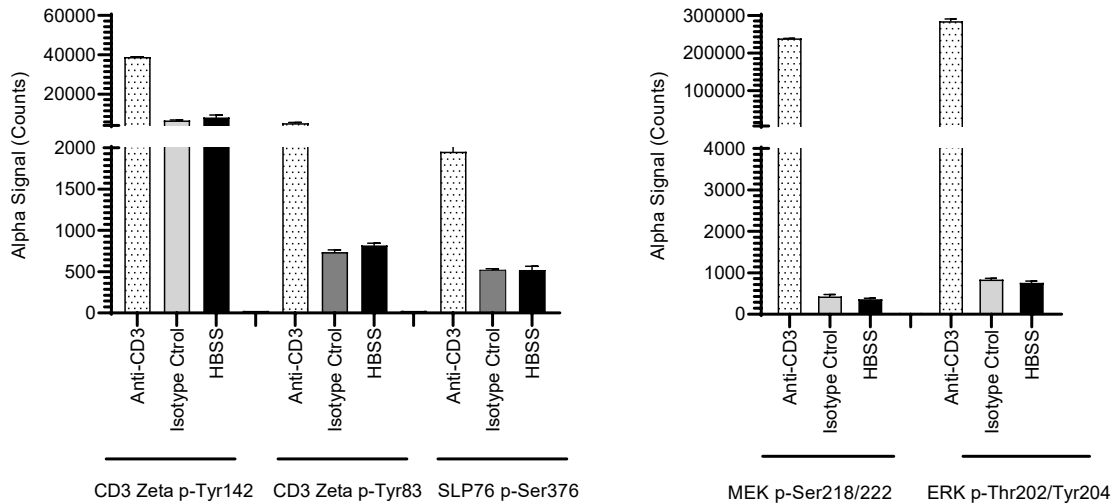
Positive Control Lysate: Prepared from Jurkat cells cultured in 10% FBS containing media. Cells were harvested and washed with HBSS + 0.1% BSA. Cells were adjusted to 3.5x10<sup>6</sup> cells/mL and treated with 44 mM H<sub>2</sub>O<sub>2</sub> for 10 minutes. Cells were then lysed with the addition of 5X Lysis Buffer.

**Representative data**

Jurkat cells were plated at 200K cells/well in a 96 well plate in HBSS + 0.1% BSA, then treated with H<sub>2</sub>O<sub>2</sub> at the indicated concentrations for 10 minutes at 37°C. Cells were lysed with 5X Lysis Buffer and assayed separately for Phospho (Tyr142) and Total CD3 Zeta using respective *SureFire Ultra* kits. Equivalent to approximately 2,000 cells/datapoint.



Data obtained with a 2-plate, 2-incubation protocol. Jurkat cells were plated at  $5 \times 10^6$  cells/mL in 12 well plates in HBSS + 0.1% BSA. Cells were treated for 10 minutes with 10  $\mu$ g/mL of anti-CD3 Ab (BD cat# 555329) or a mouse IgG isotype control. Cells were lysed with the addition of 5X Lysis Buffer and lysates were further diluted in Lysis Buffer before assayed for various targets. Equivalent to approximately 8,000 cells/datapoint.



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