AlphaLISA™ *SureFire*® Biotin Free

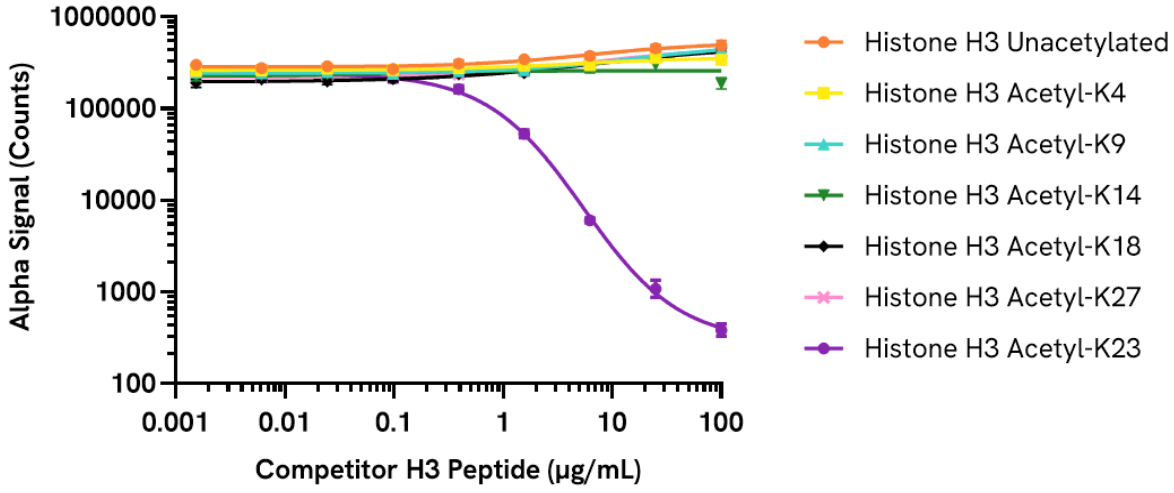
Human and Mouse Acetyl Histone H3 (K23) Detection Kit

Product number: ASBF-EHISH3-C500, ASBF-EHISH3-C10K, ASBF-EHISH3-C50K, ASBF-EHISH3-C-HV

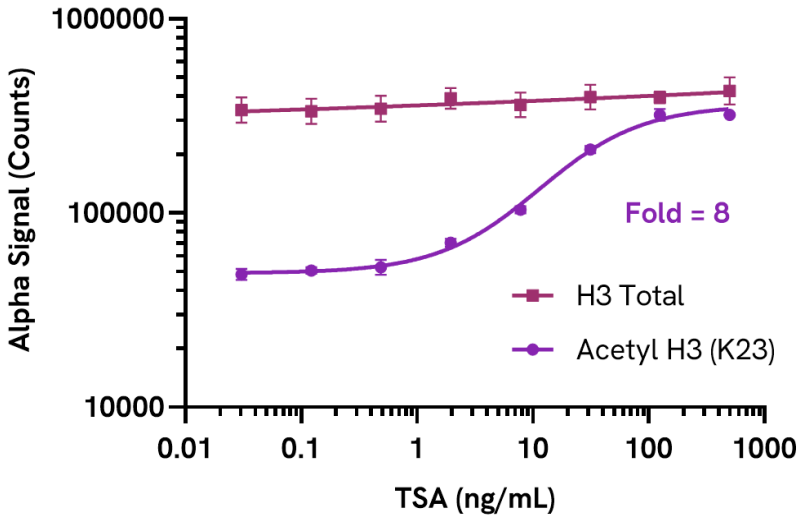
Kit specificity:
This assay kit contains antibodies which recognize the Acetyl-Lys23 epitope and a distal epitope on Histone H3. The protein detected by this kit corresponds to UniProt ID P68431. Histone H3 is also known as Histone H3.1. These antibodies recognize Acetylated Histone H3 (K23) of human 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, then treated with 500 ng/mL Trichostatin A (TSA) for 4 hours, and lysed with 120 mL of Lysis Buffer B.

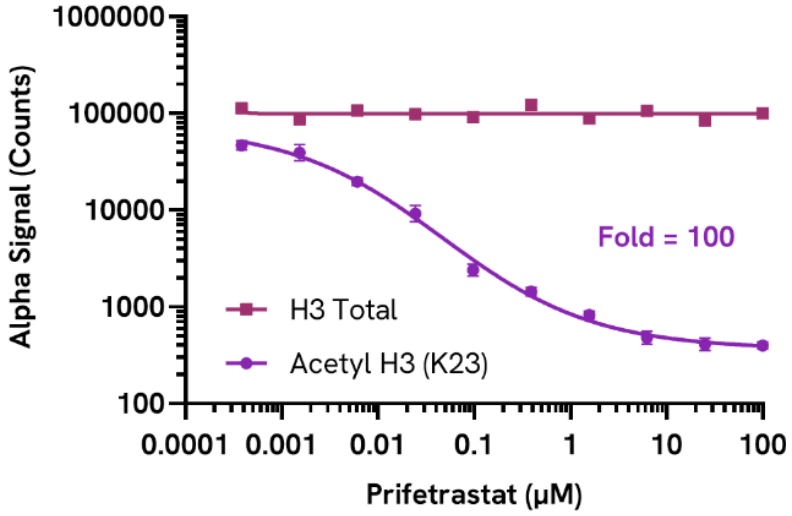
Representative data:
Specificity of the Acetyl Histone H3 (K23) assay was assessed by a peptide competition assay. Acetyl Histone H3 K4, K9, K14, K18, K23, K27 and non-acetyl peptides were serially diluted into a fixed concentration of HeLa positive control lysate. Lysates were then assayed using the Acetyl Histone H3 (K23) *SureFire* Biotin Free kit.



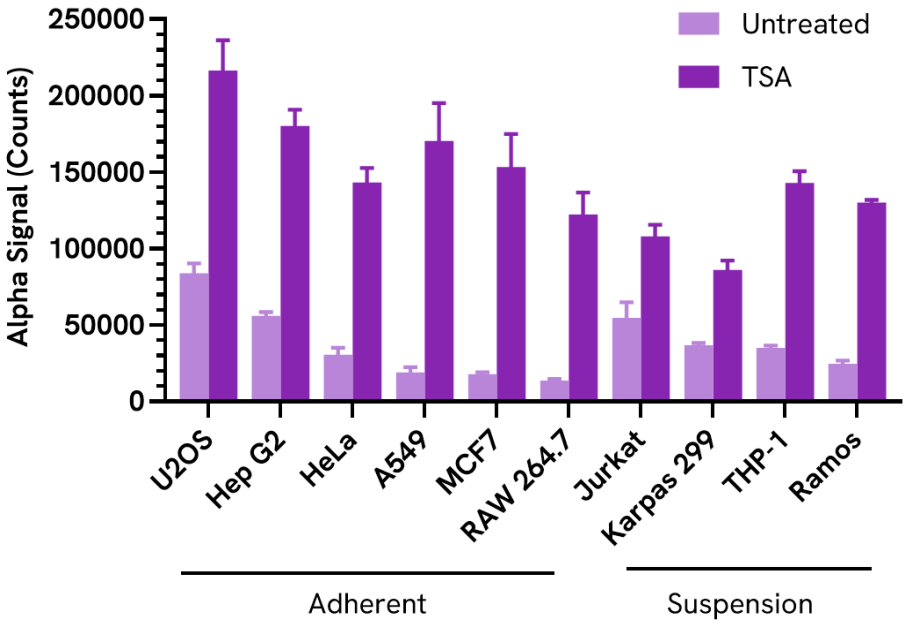
Data obtained with a 2-plate, 2-incubation protocol. HeLa cells were seeded at 200K cells/well in a 96-well plate and treated with increasing concentrations of TSA for 4 hours. Cells were lysed with 200 µL Lysis Buffer B for 30 minutes with shaking. Lysate was further diluted with Lysis Buffer B and assayed separately for Acetyl Histone H3 (K23) and Histone H3 Total using respective *SureFire* Biotin Free kits. Equivalent to approximately 200 cells/datapoint for Acetyl Histone H3 (K23) and 100 cells/datapoint for H3 Total.



Data obtained with a 2-plate, 2-incubation protocol. MCF7 cells were seeded at 40K cells/well in a 96-well plate and treated with increasing concentrations of Prifetrastat for 2 hours. Cells were lysed with 100 µL Lysis Buffer B for 30 minutes with shaking. Lysate was further diluted with Lysis Buffer B and assayed separately for Acetyl Histone H3 (K23) and Histone H3 Total using respective *SureFire* Biotin Free kits. Equivalent to approximately 400 cells/datapoint for Acetyl Histone H3 (K23) and 100 cells/datapoint for H3 Total.



Data obtained from measurement of Acetyl Histone H3 (K23) in various cell types treated with 500 ng/mL TSA for 4 hours and lysed with Lysis Buffer B for 30 minutes with shaking. Equivalent to approximately 500 cells/datapoint for adherent cells or 1,000 cells/datapoint for suspension cells.



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