

Human/Mouse Histone Deacetylase 8/HDAC8 Antibody

Antigen Affinity-purified Polyclonal Sheep IgG Catalog Number: AF4359

DESCRIPTION			
Species Reactivity	Human/Mouse		
Specificity	Detects human HDAC8 in direct ELISAs and Western blots.		
Source	Polyclonal Sheep IgG		
Purification	Antigen Affinity-purified		
Immunogen	S. frugiperda insect ovarian cell line Sf 21-derived recombinant human HDAC8 isoform 1 Met1-Val377 (Leu31Pro) Accession # Q9BY41		
Formulation	Lyophilized from a 0.2 µm filtered solution in PBS with Trehalose. See Certificate of Analysis for details. *Small pack size (-SP) is supplied either lyophilized or as a 0.2 µm filtered solution in PBS.		

APPLICATIONS

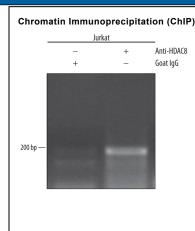
Please Note: Optimal dilutions should be determined by each laboratory for each application. General Protocols are available in the Technical Information section on our website.

	Recommended Concentration	Sample
Western Blot	1 μg/mL	See Below
Chromatin Immunoprecipitation (ChIP)	5 μg/5 x 10 ⁶ cells	See Below

DATA

| KDa | SH | HDAC8 | H

Detection of Human Histone Deacetylase 8/HDAC8 by Western Blot. Western blot shows lysates of HEK293 human embryonic kidney cell line. PVDF membrane was probed with 1 µg/mL of Sheep Anti-Human/Mouse Histone Deacetylase 8/HDAC8 Antigen Affinity-purified Polyclonal Antibody (Catalog # AF4359) followed by HRP-conjugated Anti-Sheep IgG Secondary Antibody (Catalog # HAF016). A specific band was detected for Histone Deacetylase 8/HDAC8 at approximately 50 kDa (as indicated). This experiment was conducted under reducing conditions and using Immunoblot Buffer Group 8.



Detection of Histone Deacetylase 8/ **HDAC8-regulated Genes by** Chromatin Immunoprecipitation. Jurkat human acute T cell leukemia cell line treated with 50 ng/mL PMA and 200 ng/mL calcium ionomycin for 30 minutes was fixed using formaldehyde, resuspended in lysis buffer, and sonicated to shear chromatin. Histone Deacetylase 8/HDAC8/DNA complexes were immunoprecipitated using 5 µg Sheep Anti-Human/Mouse Histone Deacetylase 8/HDAC8 Antigen Affinitypurified Polyclonal Antibody (Catalog # AF4359) or control antibody (Catalog # 5-001-A) for 15 minutes in an ultrasonic bath, followed by Biotinylated Anti-Sheep IgG Secondary Antibody (Catalog # BAF016). Immunocomplexes were captured using 50 µL of MagCellect Streptavidin Ferrofluid (Catalog # MAG999) and DNA was purified using chelating resin solution. The IFN-v promoter was detected by standard PCR.

PREPARATION AND STORAGE

Reconstitution Reconstitute at 0.2 mg/mL in sterile PBS

Shipping The product is shipped at ambient temperature. Upon receipt, store it immediately at the temperature recommended below.

*Small pack size (-SP) is shipped with polar packs. Upon receipt, store it immediately at -20 to -70 °C

- 12 months from date of receipt, -20 to -70 °C as supplied.
- 1 month, 2 to 8 °C under sterile conditions after reconstitution.
- 6 months, -20 to -70 °C under sterile conditions after reconstitution

BACKGROUND

HDAC8 (Histone deacetylase 8) is a 50 kDa member of the class I subfamily, histone deacetylase family of transcriptional regulators. It is a smooth muscle enzyme that deacetylates histone lysines, blocking gene transcription. HDAC8 appears to function as a monomer, and phosphorylation at Ser39 inhibits activity. Human HDAC8 is 377 amino acids (aa) in length. It contains a series of α helices and strands, plus a catalytic ferrous ion binding site (Asp178-His180-Asp267). There are four potential splice variants, all of which possess an alternate start site 38 aa upstream of the standard start site. Three have additional changes; two show a one and three aa substitution respectively for aa 211-377, while a third has a 15 aa substitution for aa 148-377. Human HDAC8 shares more than 96% aa identity with both mouse and canine HDAC8.

Rev. 2/6/2018 Page 1 of 1

