Acetylation and deacetylation dynamics Antibody Sampler Kit

HAK21160

Contains Product	Quantity	Applications	Species reactivity	MW(kDa)
HDAC1 [ET1605-35]	20µl	WB,IHC-P,IF-Cell,IP,IF-Tissue	H,M,R	55 kDa
HDAC3 [ET1610-5]	20µl	WB,IF-Cell,IF-Tissue,IHC-P,IP	H,M,R	49 kDa
SIRT1 [ET1603-3]	20µl	WB,IF-Cell,IF-Tissue,IHC-P,ChIP	H,M,R	82 kDa
GCN5 [ER63516]	20µl	WB,IHC-P	H,M	94 kDa
KAT7 [ET1705-25]	20µl	WB,IHC-P,IP,IF-Cell	H,M,R	71 kDa
HRP-Goat Anti-Rabbit IgG (H+L) [HA1001]	100µl	WB,ELISA,IHC-P	Rab	

Description: The Acetylation and deacetylation dynamics Antibody Sampler Kit provides a fast and economical means of evaluating the acetylation and deacetylation. The kit contains enough primary and secondary antibodies to perform two Western mini-blot experiments.

Storage Buffer: 1*TBS (pH7.4), 0.05% BSA, 40% Glycerol. Preservative: 0.05% Sodium Azide.

Storage Instruction: Store at +4℃ after thawing. Aliquot store at -20℃. Avoid repeated freeze / thaw cycles.

Background Epigenetic regulators, particularly those modulating histone acetylation, play a pivotal role in controlling gene expression and cellular processes. This balance is maintained by opposing actions: histone acetyltransferases (HATs), such as GCN5 and KAT7, add acetyl groups to lysine residues on histones, promoting chromatin relaxation and transcriptional activation. In contrast, histone deacetylases (HDACs), including HDAC1, HDAC3, and SIRT1, remove these groups, leading to chromatin condensation and gene repression.

These enzymes are crucial for normal development, and their dysregulation is implicated in various diseases, making them significant therapeutic targets. Thurs,these targets represent a powerful network for epigenetic intervention in diseases like cancer and metabolic disorders.

Database links: UniProt ID: Q13547, O09106, Q4QQW4, O15379, O88895, Q6P6W3, Q96EB6, Q923E4, 309757, Q92830, O95251, Q5SVQ0, Q810T5

Images

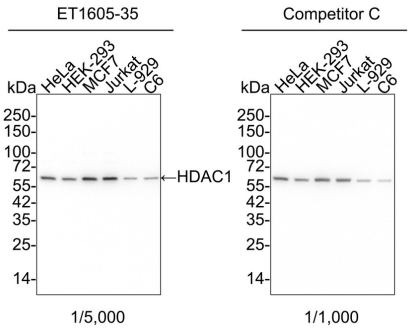

Fig1: Western blot analysis of HDAC1 on different lysates with Rabbit anti-HDAC1 antibody (ET1605-35) at 1/5,000 dilution and competitor's antibody at 1/1,000 dilution.

Lane 1: HeLa cell lysate
Lane 2: HEK-293 cell lysate
Lane 3: MCF7 cell lysate
Lane 4: Jurkat cell lysate
Lane 5: L-929 cell lysate
Lane 6: C6 cell lysate

Lysates/proteins at 15 µg/Lane.

Predicted band size: 55 kDa
Observed band size: 65 kDa

Exposure time: 30 seconds; ECL: K1801;
4-20% SDS-PAGE gel.

Proteins were transferred to a PVDF membrane and blocked with 5% NFDM/TBST for 1 hour at room temperature. The primary antibody (ET1605-35) at 1/5,000 dilution and competitor's antibody at 1/1,000 dilution were used in 5% BSA at 4℃ overnight. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1/50,000 dilution was used for 1 hour at room temperature.

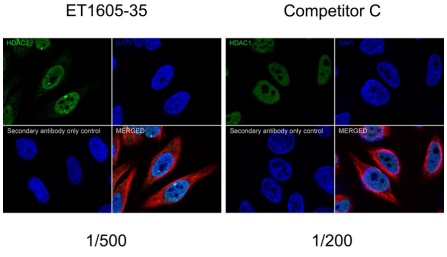

Fig2: Immunocytochemistry analysis of HeLa cells labeling HDAC1 with Rabbit anti-HDAC1 antibody (ET1605-35) at 1/500 dilution and competitor's antibody at 1/200 dilution.

Cells were fixed in 4% paraformaldehyde for 20 minutes at room temperature, permeabilized with 0.1% Triton X-100 in PBS for 5 minutes at room temperature, then blocked with 1% BSA in 10% negative goat serum for 1 hour at room temperature. Cells were then incubated with Rabbit anti-HDAC1 antibody (ET1605-35) at 1/500 dilution and competitor's antibody at 1/200 dilution in 1% BSA in PBST overnight at 4 ℃. Goat Anti-Rabbit IgG H&L (iFluor™ 488, HA1121) was used as the secondary antibody at 1/1,000 dilution. PBS instead of the primary antibody was used as the secondary antibody only control. Nuclear DNA was labelled in blue with DAPI.

Beta tubulin (M1305-2, red) was stained at 1/100 dilution overnight at +4℃. Goat Anti-Mouse IgG H&L (iFluor™ 594, HA1126) was used as the secondary antibody at 1/1,000 dilution.

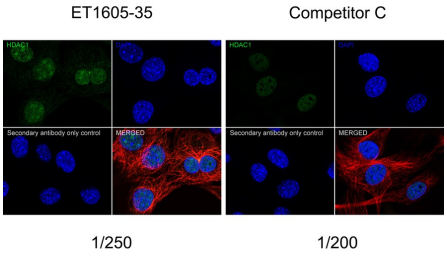

Fig3: Immunocytochemistry analysis of NIH/3T3 cells labeling HDAC1 with Rabbit anti-HDAC1 antibody (ET1605-35) at 1/250 dilution and competitor's antibody at 1/200 dilution.

Cells were fixed in 4% paraformaldehyde for 20 minutes at room temperature, permeabilized with 0.1% Triton X-100 in PBS for 5 minutes at room temperature, then blocked with 1% BSA in 10% negative goat serum for 1 hour at room temperature. Cells were then incubated with Rabbit anti-HDAC1 antibody (ET1605-35) at 1/250 dilution and competitor's antibody at 1/200 dilution in 1% BSA in PBST overnight at 4 ℃. Goat Anti-Rabbit IgG H&L (iFluor™ 488, HA1121) was used as the secondary antibody at 1/1,000 dilution. PBS instead of the primary antibody was used as the secondary antibody only control. Nuclear DNA was labelled in blue with DAPI.

Beta tubulin (M1305-2, red) was stained at 1/100 dilution overnight at +4℃. Goat Anti-Mouse IgG H&L (iFluor™ 594, HA1126) was used as the secondary antibody at 1/1,000 dilution.

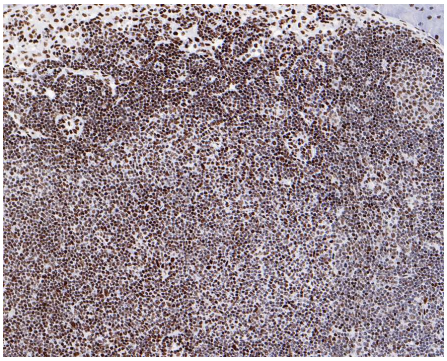

Fig4: Immunohistochemical analysis of paraffin-embedded mouse lymph nodes tissue with Rabbit anti-HDAC1 antibody (ET1605-35) at 1/800 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) (high pressure) for 2 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (ET1605-35) at 1/800 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

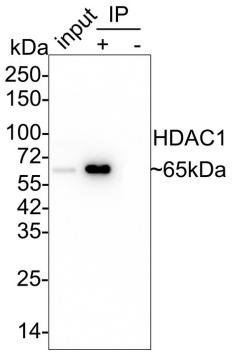

Fig5: HDAC1 was immunoprecipitated in 0.2mg HeLa cell lysate with ET1605-35 at 2 µg/10 µl beads. Western blot was performed from the immunoprecipitate using ET1605-35 at 1/5,000 dilution. Anti-Rabbit IgG for IP Nano-secondary antibody (NBI01H) at 1/5,000 dilution was used for 1 hour at room temperature.

Lane 1: HeLa cell lysate (input)
Lane 2: ET1605-35 IP in HeLa cell lysate
Lane 3: Rabbit IgG instead of ET1605-35 in HeLa cell lysate

Blocking/Dilution buffer: 5% NFDM/TBST
Exposure time: 43 seconds; ECL: K1802

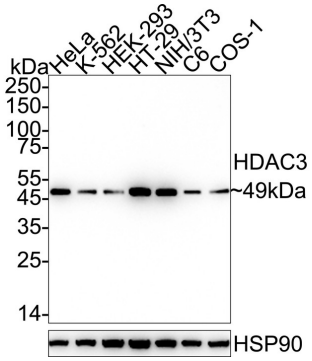

Fig6: Western blot analysis of HDAC3 on different lysates with Rabbit anti-HDAC3 antibody (ET1610-5) at 1/2,000 dilution.

Lane 1: HeLa cell lysate
Lane 2: K-562 cell lysate
Lane 3: HEK-293 cell lysate
Lane 4: HT-29 cell lysate
Lane 5: NIH/3T3 cell lysate
Lane 6: C6 cell lysate
Lane 7: COS-1 cell lysate

Lysates/proteins at 20 µg/Lane.

Predicted band size: 49 kDa
Observed band size: 49 kDa

Exposure time: 3 minutes; ECL: K1801;

4-20% SDS-PAGE gel.

Proteins were transferred to a PVDF membrane and blocked with 5% NFDM/TBST for 1 hour at room temperature. The primary antibody (ET1610-5) at 1/2,000 dilution was used in 5% NFDM/TBST at 4°C overnight. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1/50,000 dilution was used for 1 hour at room temperature.

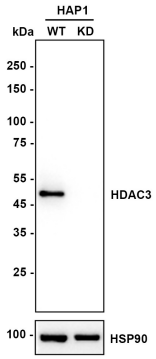

Fig7: Western blot analysis of HDAC3 on different lysates with Rabbit anti-HDAC3 antibody (ET1610-5) at 1/5,000 dilution.

Lane 1: HAP1-parental cell lysate
Lane 2: HAP1-HDAC3 KD cell lysate

Lysates/proteins at 10 µg/Lane.

Predicted band size: 49 kDa
Observed band size: 49 kDa

Exposure time: 30 seconds; ECL: K1801;

4-20% SDS-PAGE gel.

Proteins were transferred to a PVDF membrane and blocked with 5% NFDM/TBST for 1 hour at room temperature. The primary antibody (ET1610-5) at 1/5,000 dilution was used in K1803 at 4℃ overnight. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1/50,000 dilution was used for 1 hour at room temperature.

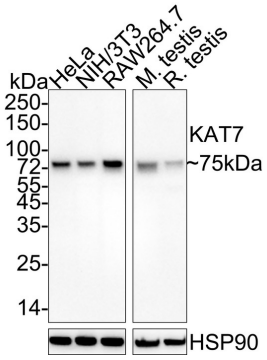

Fig8: Western blot analysis of KAT7 on different lysates with Rabbit anti-KAT7 antibody (ET1705-25) at 1/1,000 dilution.

Lane 1: HeLa cell lysate (20 µg/Lane)
Lane 2: NIH/3T3 cell lysate (20 µg/Lane)
Lane 3: RAW264.7 cell lysate (20 µg/Lane)
Lane 4: Mouse testis tissue lysate (40 µg/Lane)
Lane 5: Rat testis tissue lysate (40 µg/Lane)

Predicted band size: 71 kDa
Observed band size: 75 kDa

Exposure time: 3 minutes;

4-20% SDS-PAGE gel.

Proteins were transferred to a PVDF membrane and blocked with 5% NFDM/TBST for 1 hour at room temperature. The primary antibody (ET1705-25) at 1/1,000 dilution was used in 5% NFDM/TBST at 4℃ overnight. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1/50,000 dilution was used for 1 hour at room temperature.

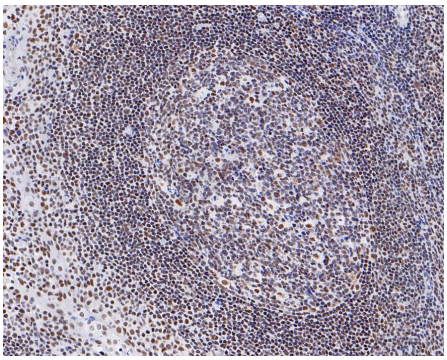

Fig9: Immunohistochemical analysis of paraffin-embedded human tonsil tissue with Rabbit anti-HDAC3 antibody (ET1610-5) at 1/200 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) (high pressure) for 2 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (ET1610-5) at 1/200 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

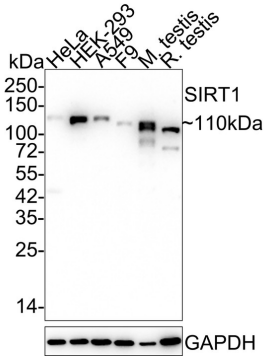

Fig10: Western blot analysis of SIRT1 on different lysates with Rabbit anti-SIRT1 antibody (ET1603-3) at 1/2,000 dilution.

Lane 1: HeLa cell lysate (15 µg/Lane)
Lane 2: HEK-293 cell lysate (15 µg/Lane)
Lane 3: A549 cell lysate (15 µg/Lane)
Lane 4: F9 cell lysate (15 µg/Lane)
Lane 5: Mouse testis tissue lysate (20 µg/Lane)
Lane 6: Rat testis tissue lysate (20 µg/Lane)

Predicted band size: 82 kDa
Observed band size: 110 kDa

Exposure time: 2 minutes 37 seconds;

4-20% SDS-PAGE gel.

Proteins were transferred to a PVDF membrane and blocked with 5% NFDM/TBST for 1 hour at room temperature. The primary antibody (ET1603-3) at 1/2,000 dilution was used in 5% NFDM/TBST at 4℃ overnight. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1/50,000 dilution was used for 1 hour at room temperature.

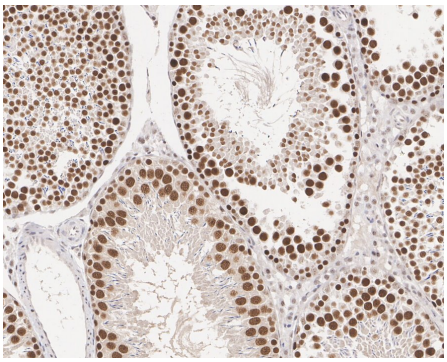

Fig11: Immunohistochemical analysis of paraffin-embedded rat testis tissue with Rabbit anti-SIRT1 antibody (ET1603-3) at 1/1,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) (high pressure) for 2 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (ET1603-3) at 1/1,000 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

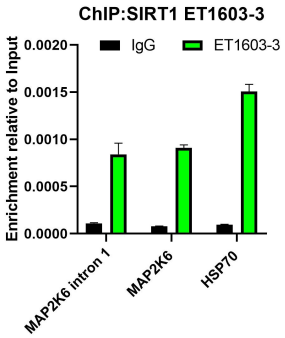

Fig12: Chromatin immunoprecipitations were performed with cross-linked chromatin from HeLa cells with SIRT1 (ET1603-3) or Normal Rabbit IgG according to the ChIP protocol. The enriched DNA was quantified by real-time PCR using indicated primers. The amount of immunoprecipitated DNA in each sample is represented as signal relative to the total amount of input chromatin, which is equivalent to one.

Note: All products are "FOR RESEARCH USE ONLY AND ARE NOT INTENDED FOR DIAGNOSTIC OR THERAPEUTIC USE".

Background References

1. Zhou MM, Cole PA. Targeting lysine acetylation readers and writers. Nat Rev Drug Discov. 2025 Feb;24(2):112-133.
2. Peterson CL, Laniel MA. Histones and histone modifications. Curr Biol. 2004 Jul 27;14(14):R546-51.
3. Shvedunova M, Akhtar A. Modulation of cellular processes by histone and non-histone protein acetylation. Nat Rev Mol Cell Biol. 2022 May;23(5):329-349.
4. Drazic A, Myklebust LM, Ree R, Arnesen T. The world of protein acetylation. Biochim Biophys Acta. 2016 Oct;1864(10):1372-401.