

# Certificate of Analysis

## HAT1

**Human Histone Acetyltransferase 1**  
(Recombinant enzyme expressed in *E.coli*)

Item # EPI025

Lot # 139718

**Product Description:** N-terminal 6His-tagged, recombinant human HAT1, amino acids 20-341, expressed in *E.coli*. Purified using immobilised metal affinity chromatography. MW = 39.8kDa.

**Alias:** KAT1

**Formulation:** 1mg/ml of enzyme in 25mM Tris/HCl pH7.5, 125mM NaCl, 0.3mM DTT, 50% Glycerol. Frozen solution.

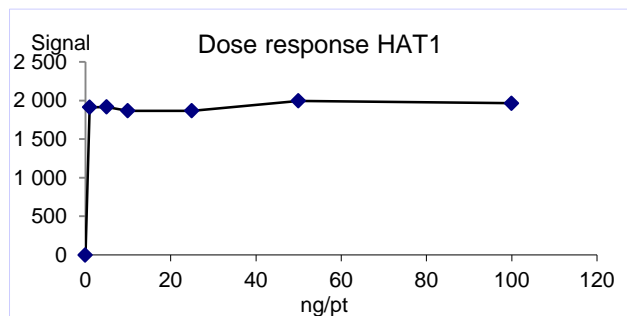
**Storage and Stability:** Stable for 1 year at -70°C from date of shipment. For maximum recovery of product, centrifuge original vial prior to removing the cap.

**Handling Recommendations:** Rapidly thaw the vial under cold water and immediately place on ice. Aliquot unused material into pre-chilled microcentrifuge tubes and store at -70°C.

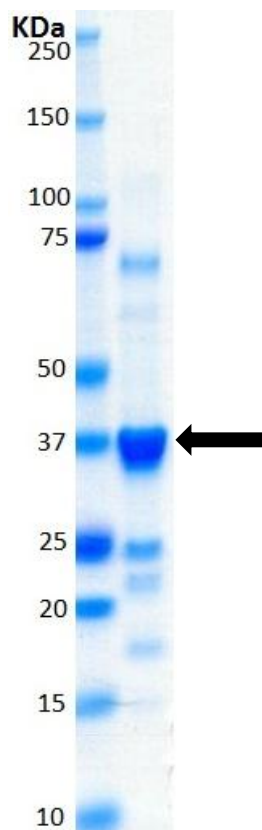
**FOR IN VITRO RESEARCH USE ONLY**  
**NOT FOR USE IN HUMANS OR ANIMALS**

## Quality Control Testing

**HAT Assay:** 1-100ng of this lot of enzyme transferred acetyl groups from [3H] Acetyl CoA to histone H4 in the assay described on page two. Assay background was subtracted from the actual counts to yield the results shown below.



**MS:** Size was confirmed by mass spectrometry using a Q-TOF.



**SDS-PAGE and Coomassie Stain:** Purity was assessed by SDS-PAGE and Coomassie blue staining using 4µg of active HAT1.

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### HAT1 Assay Protocol

#### Stock Solutions:

1. **Reaction buffer:** 50mM Hepes/Tris pH=7.4, 0.1mM EDTA/Tris, 0.01% Triton.
2. **HAT1:** Dilute with reaction buffer. Use 1-100ng per assay point.
3. **Histone H4 full length human:** Dilute with reaction buffer to 750nM.
4. **[3H] Acetyl Coenzyme A:** Dilute with reaction buffer to 6nM.
5. **Filtration Buffer :** 33mM Citric acid pH2.2

#### Assay Procedure (96 well plate format):

1. Add 5 $\mu$ l of 10% DMSO per assay to wells.
2. Add 25 $\mu$ l of [3H] Acetyl Coenzyme A.
3. Add 10 $\mu$ l (**1-100ng**) HAT1.
4. Add 10 $\mu$ l of Histone H4 full length human.
5. Incubate for 10 minutes at 22°C.
6. Stop the reaction by adding 500 $\mu$ l of citric acid, then filter on a GF/B Filter. Wash 3 times with Filtration buffer.
7. Dry and add scintillation cocktail.
8. Read in a scintillation counter. Compare the signal of enzyme samples with that of background sample that contain all assay components except the enzyme HAT1.

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### HAT1 Sequence Information

**Protein** Human HAT1  
**Tags** N-terminal 6His  
**Accession number** NP\_003633.1

***Recombinant HAT1 amino acid sequence:***

1 MGSSHHHHHH SSGLVPRGSK KLAEYKCNTN TAIELKLVRF PEDLENDIRT  
51 FFPEYTHQLF GDETAFGYK GLKILLYYIA GSLSTMFRVE YASKVDENFD  
101 CVEADDDVEGK IRQIIPPGFC TNTNDFLSLL EKEVDFKPFQ TLLHTYSVLS  
151 PTGGENFTFQ IYKADMTCRG FREYHERLQT FLMWFIETAS FIDVDDERWH  
201 YFLVFEKYNK DGATLFATVG YMTVYNYVVY PDKTRPRVSQ MLILTPFQGQ  
251 GHGAQLLETV HRYYTEFPTV LDITAEDPSK SYVKLRDFVL VKLCQDLPCF  
301 SREKLMQGFN EDMAIEAQQK FKINKQHARR VYEILRLLVT D

Reviewed and approved by site quality representative.

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