

Certificate of Analysis

SETD7

Human histone Lysine N-methyltransferase SETD7, active

(Recombinant enzyme expressed in *E.coli*)

Item # EPI056

Lot # 139699

Product Description: C-terminal, 6His-tagged recombinant human SETD7, amino acids 1-366, expressed in *E.coli*. Purified using immobilised metal affinity chromatography. MW = 41.5kDa.

Formulation: 1mg/ml of enzyme in 5mM Tris/HCl pH7.5, 250mM NaCl, 65% Glycerol, 0.6mM TCEP. Frozen solution.

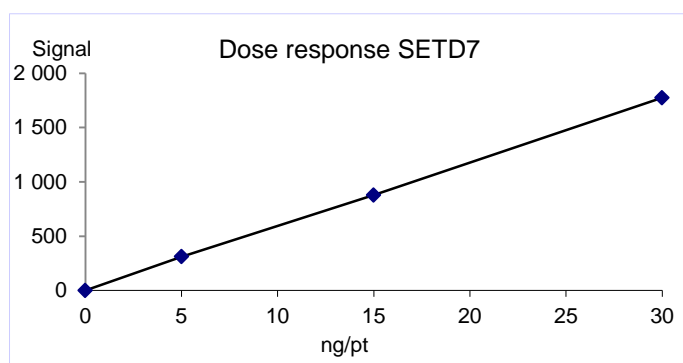
Storage and Stability: On receipt of material store at -70°C. Unopened reagent is stable for a minimum of 1 year from date of shipment when stored at recommended storage temperature. Avoid repeat freeze/thaw cycles. For maximum recovery of product, centrifuge original vial prior to removing the cap.

Handling Recommendations: Rapidly thaw the vial under cold water and immediately place on ice. Aliquot unused material into pre-chilled micro-centrifuge tubes and immediately snap-freeze the vials in liquid nitrogen prior to re-storage at -70°C.

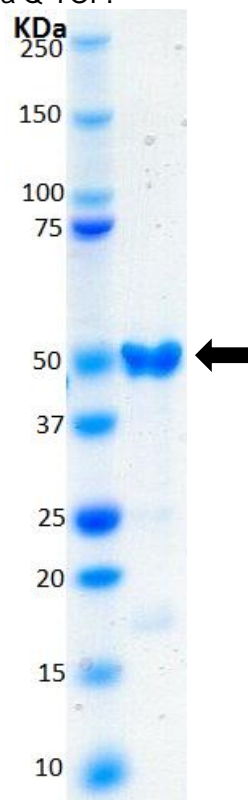
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NOT FOR USE IN HUMANS OR ANIMALS

Quality Control Testing

HMT Assay: 5-30ng of this lot of enzyme transferred methyl groups from [3H] SAM to Histone H3 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 SETD7.



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SETD7 Assay Protocol

Stock Solutions:

1. **Reaction buffer:** 50mM Tris/HCl pH9, 50mM NaCl, 5mM MgCl₂, 4mM DTT.
2. **SETD7, active:** Dilute with reaction buffer. Use 5-30ng per assay point.
3. **H3 Histone:** Dilute with reaction buffer to 600nM.
4. **[3H] SAM:** Dilute with SAM solution (1530nM) to 170nM.
5. **Filtration Buffer :** 33mM Citric acid pH2.2

Assay Procedure (96 well plate format):

1. Add 5µl of 10% DMSO per assay to each well.
2. Add 25µl of [3H] SAM.
3. Add 10µl **(5-30ng) SETD7, active**.
4. Add 10µl of Histone H3.
5. Incubate for 30 minutes at 22°C.
6. Stop the reaction by adding 500µl 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 a background sample that contains all assay components except the enzyme SETD7.

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SETD7 Sequence Information

Protein Human SETD7
Tags C-Terminal 6His
Accession number GenBank NP_085151.1

Recombinant SETD7 amino acid sequence:

1 MDSDDDEMVVEE AVEGHLDDDG LPHGFCTVTY SSTDRFEGNF VHGEKNGRGK
51 FFFFDGSTLE GYYVDDALQG QGVYTYEDGG VLQGTYYVDGE LNGPAQEYDT
101 DGRILIFKGQY KDNIRHGVWCW IYYPDGGSLV GEVNEDGEMT GEKIAYVYPD
151 ERTALYGKFI DGEMIEGKLA TLMSTEEGRP HFELMPGNSV YHFDKSTSSC
201 ISTNALLPDP YESERVYVAE SLISSAGEGL FSKVAVGPNT VMSFYNGVRI
251 THQEVDSDRW ALNGNTLSLD EETVIDVPEP YNHVSKYCAS LGHKANHSFT
301 PNCIYDMFVH PRFGPIKCIR TLRAVEADEE LTVAYGYDHS PPGKSGPEAP
351 EWYQVELKAF QATQQKHHHH HH

Reviewed and approved by site quality representative.

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