

SMYD2

(SET and MYND Domain-Containing Protein-2; Lysine N-Methyltransferase 3C; KMT3C)

CATALOG NO.: HMT-11-110

LOT NO.:

DESCRIPTION: Human recombinant SMYD2 (residues 2-433 (C-terminus); Genbank Accession # NM_020197) expressed with an N-terminal His-tag, in *E. coli*. MW = 52.0 kDa. Catalyzes the transfer of methyl groups from S-adenosyl-L-methionine (SAM) to the ε-amino function of protein L-lysine residues, particularly in histones H4 and H3, although other activities have been reported (see below). Lysine methyltransferases of the SMYD family are unusual in that their SET domains are split in two by insertion of a myeloid-Nervy-DEAF-1 (MYND) domain in the primary amino acid sequence¹⁻³. SMYD2 is widely expressed, but at the highest levels in heart and brain tissues^{1,3,4}. SMYD2 is frequently overexpressed in esophageal squamous cell carcinoma (ESCC) and SMYD2 knockdown inhibits the proliferation of SMYD2-overexpressing ESCC cell lines⁵. Although SMYD2 has been described as a histone H3 (H3K4⁶, H3K36³), p53 (p53K370)⁷ and Rb (retinoblastoma; RbK860)⁸ lysine methyltransferase, RBC finds this recombinant SMYD2 preparation to be far more active with recombinant histone H4 than with histone H3, an H3(1-21) peptide, an H4(1-21) peptide, core histones or recombinant p53. Similar results have been reported for both human recombinant SMYD2⁹ and SMYD3¹⁰, with the methylation site, in the SMYD3 case, identified as H4K20¹⁰.

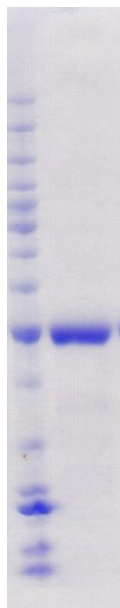
PURITY: >90% by SDS-PAGE.

ASSAY CONDITIONS: RBC's SMYD2 displays histone methyltransferase activity at enzyme concentrations of 26 nM and above, 30°C, with recombinant human histone H4 or calf thymus histone H3 in the HMT HotSpotSM Assay format. Reaction conditions are: 50 mM Tris-HCl, pH 8.5, 50 mM NaCl, 5 mM MgCl₂, 1 mM DTT, 1 mM PMSF, histone H4 (5 μM) or histone H3 (5 μM), [³H]-SAM.

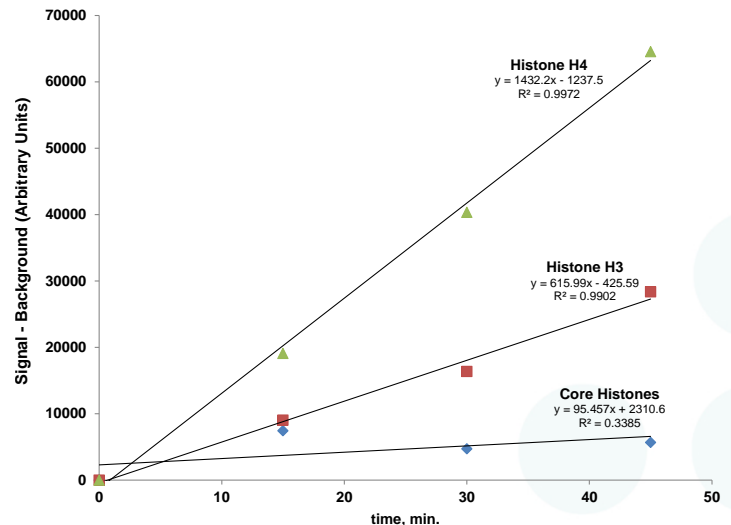
SUPPLIED AS: ___ μg/μl in 20 mM Tris/HCl, pH 7.0, 120 mM NaCl, 1 mM TCEP, 1 μM ZnCl₂, 10% (w/v) glycerol as determined by OD₂₈₀

STORAGE: -70°C. Thaw quickly and store on ice before use. The remaining, unused, undiluted enzyme should be refrozen quickly by, for example, snap freezing in a dry/ice ethanol bath or liquid nitrogen. Freezing and storage of diluted enzyme is not recommended.

REFERENCES: 1) P.D. Gottlieb *et al. Nat. Genet.* 2002 **31** 25; 2) R. Hamamoto *et al. Nat. Cell Biol.* 2004 **6** 731; 3) M.A. Brown *et al. Mol. Cancer* 2006 **5** 26; 4) I. Hwang & P.D. Gottlieb *J. Immunol.* 1997 **158** 1165 5) S. Komatsu *et al. Carcinogenesis* 2009 **30** 1139; 6) M. Abu-Farha *et al. Mol. Cell. Proteomics* 2008 **7** 560; 7) J. Huang *et al. Nature* 2006 **444** 629; 8) L.A. Saddic *et al. J. Biol. Chem.* 2010 **285** 37733; 9) J. Wu *et al. Biochemistry* 2011 **50** 6488; 10) K.W. Foreman *et al. PLoS one* 2011 **6** e22290



Coomassie blue stained SDS-PAGE (4-12% acrylamide) of 4 μg of purified RBC SMYD2. MW markers (left) are, from top, 220, 160, 120, 90, 70, 60, 50, 40, 30, 25, 20, 15, 10 kDa.



Time courses of SMYD2 methyltransferase reactions in the HotSpotSM assay format. SMYD2, at 58.5 nM, was assayed with 0.05 mg/mL chicken core histones, 5 μM histone H3 or 5 μM histone H4, plus 1 μM [³H]-SAM. Points represent the mean of two determinations and lines/equations derive from linear least-squares fits.

This product is not intended for therapeutic or diagnostic use in animals or in humans.