

PRMT5

(Protein Arginine Methyltransferase 5)

CATALOG NO.: HMT-21-172

LOT NO.:

DESCRIPTION: Full-length human recombinant PRMT5 expressed in *Sf9* insect cells (residues 2-637 (C-terminus); Genbank Accession # NM_006109; N-terminal Flag-tag; MW = 73.7). PRMT5, a type II arginine methyltransferase, catalyzes the transfer of a methyl group from S-adenosyl-L-methionine (SAM) to an ω -nitrogen of the guanidino function of protein L-arginine residues (ω -monomethylation) and the transfer of a second methyl group to the other ω -nitrogen, yielding symmetric dimethylarginine (sDMA)¹. PRMT5 catalytic activity with certain substrates (e.g. histones H2A, H4) can be weak when PRMT5 is not complexed with MEP50, which may be due to MEP50's role in substrate binding³. However, Reaction Biology has found that PRMT5 alone displays significant activity with GST-GAR (see Figure below). The GAR domain of this substrate ("glycine and arginine rich") derives from the N-terminus of fibrillarin, a nucleolar protein and RNA methyltransferase that can form a protein complex whose components include PRMT5 and PRMT1³. In complex with MEP50, PRMT5 is also a component of multiple macromolecular complexes (e.g. 20S Methylosome, Swi/Snf), is located in both the nucleus and cytoplasm, modifies a variety of substrates and plays roles in chromatin remodeling, RNA processing, and regulation of gene expression, cell growth and differentiation (see reviews^{4,5}). PRMT5's pro-proliferative effects⁶⁻⁹ and their association with multiple cancers (lung^{10,11}, breast⁹, ovarian¹², lymphoid^{7,8}) has led to increased interest in its possible targeting for anti-cancer therapy.

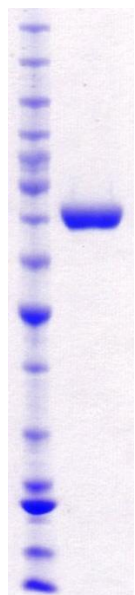
PURITY: >90% by SDS-PAGE.

ASSAY CONDITIONS: RBC's PRMT5 displays substantial methyltransferase activity from [³H]-SAM to GST-GAR (Cat. #HMT-11-137; see Figure, below). Activity was determined as TCA-precipitated counts in a scintillation/filter plate assay (Multiscreen FB, Topcount). Reaction conditions: 50 mM Tris-HCl, pH 8.5, 50 mM NaCl, 5 mM MgCl₂, 1 mM DTT, 1 mM PMSF, 30°C, 60 min. with 1 μ M [³H]-SAM and 0.05 mg/mL (1.5 μ M) GST-GAR as substrates.

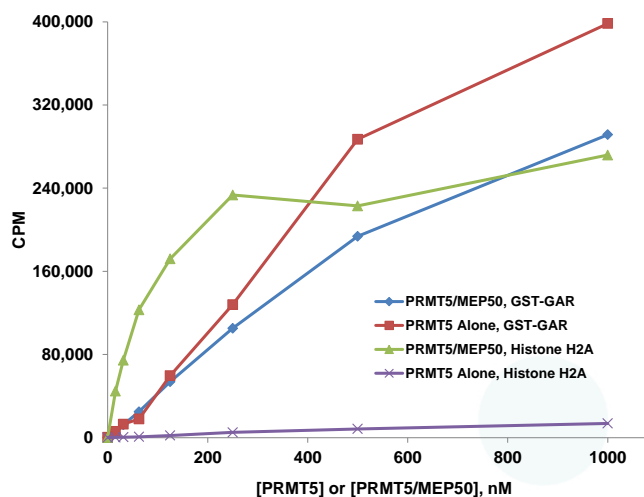
SUPPLIED AS: ___ μ g/ μ l total protein in 50 mM Tris/HCl, pH 8.0, 110 mM NaCl, 2.2 mM KCl, 3 mM TCEP, 20% (v/v) glycerol as determined by OD₂₈₀

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) T.L. Branscombe *et al. J. Biol. Chem.* 2001 **276** 32971; 2) M. Ho *et al. PLOS One* 2013 **8** e57008; 3) M. Yanagida *et al. J. Biol. Chem.* 2004 **279** 1607; 4) S.S. Wolf *Cell Mol. Life Sci.* 2009 **66** 2109; 5) V. Kharkanis *et al. Trends Biochem. Sci.* 2011 **36** 633; 6) S. Pal *et al. Mol. Cell. Biol.* 2004 **24** 9630; 7) L. Wang *et al. Mol. Cell. Biol.* 2008 **28** 6262; 8) P. Aggarwal *et al. Cancer Cell* 2010 **18** 329; 9) M.A. Powers *et al. Cancer Res.* 2011 **71** 5579; 10) T.Y. Wei *et al. Cancer Sci.* 2012 **103** 1640; 11) Z. Gu *et al. Biochem. J.* 2012 **446** 235; 12) X. Bao *et al. J. Histochem. Cytochem.* 2013 **61** 206;



Coomassie blue stained SDS-PAGE (4-12% acrylamide) of 4 μ g of RBC PRMT5. MWs of markers at left are, from top (kDa): 220, 160, 120, 100, 90, 80, 70, 60, **50**, 40, 30, 25, **20**, 15, & 10 kDa.



Methyltransferase Activities of PRMT5 alone vs. those of PRMT5/MEP50. Methylation determined as TCA-precipitable counts in a scintillation/filter plate assay. Reactions were 60 min., 30°C, with 1 μ M [³H]-SAM and either 0.05 mg/mL (1.5 μ M) GST-GAR or 1 μ M (H2A) as protein substrate.

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

Reaction Biology

1 Great Valley Parkway, Malvern PA, USA 19355

requests@reactionbiology.com www.reactionbiology.com