

Recombinant KMT2C (MLL3) complex

Catalog No: 31478, 31878

Lot No: 21815002

Expressed In: *E. coli*

Quantity: 20 µg

Concentration: 1.4 µg/µl

Source: Human

Buffer Contents: Full length Recombinant KMT2C (MLL3) complex expressed in *E. coli* cells at a concentration of 1.4 µg/µl in 25 mM Tris pH 8.0, 300 mM NaCl, 5% Glycerol, 0.04% Triton X-100.

Background: The **Recombinant KMT2C (MLL3) complex** (MLL3/WDR5/ASH2L/RBBP5/DPY30), also known as the **MLL3 WARD Complex** is a histone methyltransferase that, like MLLs 1, 2 and 4, preferentially methylates the activating histone mark lysine 4 of histone H3 (H3K4). The KMT2C (also known as MLL3) subunit contains the catalytic SET domain that is responsible for substrate recognition and enzymatic activity, along with the WDR5 and RBBP5 interaction motifs necessary for assembly of the WARD complex. The MLL3 WARD complex forms part of the ASC-2/NCOA6 complex (ASCOM), which possesses histone methylation activity and is involved in transcriptional coactivation. The chromatin remodeling complex SWI/SNF also interacts with ASCOM through binding of the MLL3 (or MLL4) SET domain with the INI1 subunit of SWI/SNF. Knockdown of MLLs 3 and 4 expression results in a reduction of HOXC10, an estrogen-induced protein that is overexpressed in breast cancer. (edit)

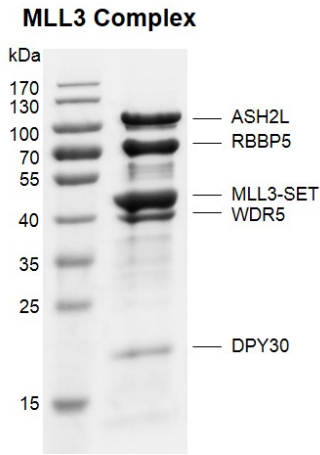
Protein Details: Recombinant KMT2C (MLL3) complex (MLL3/WDR5/ASH2L/RBBP5/DPY30) was generated using a peptide corresponding to amino acids 4740-4911 (accession number NP_733751.2) of the KMT2C (MLL3) protein that was expressed in *E. coli*. Recombinant KMT2C (MLL3) complex contains an N-terminal GST tag with a molecular weight of 46 kDa; human WDR5 (accession number NM_017588) that contains an N-terminal His-tag with a molecular weight of 35 kDa; human ASH2L (accession number NM_001105214) that contains an N-terminal His-tag with a molecular weight of 61 kDa; human RBBP5 (accession number NM_005057) that contains an N-terminal His-tag with a molecular weight of 60kDa; and human DPY30 (accession number NM_032574) that contains an N-terminal His-tag with a molecular weight of 12 kDa. All Recombinant MLL3/WDR5/ASH2L/RBBP5/DPY30 Complex proteins were individually expressed in an *E. coli* expression system. The recombinant protein is >90% pure by SDS-PAGE.

Application Notes: Recombinant KMT2C (MLL3) complex is suitable for use in the study of enzyme kinetics, inhibitor screening, and selectivity profiling.

Specific Activity: H3K4me(1-3) methyltransferase

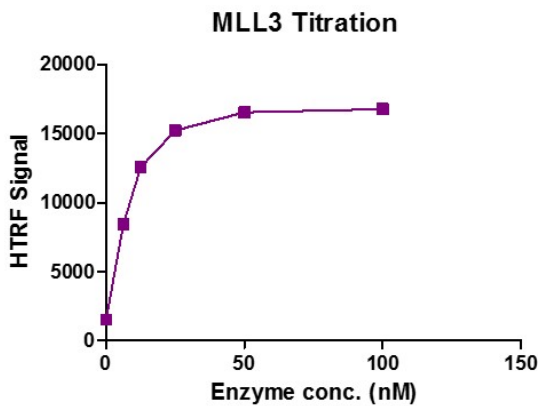
HMT Assay Conditions: 3.3 µM H3K4me0 (1-21aa) peptide was incubated with different concentrations of Recombinant KMT2C (MLL3) complex in reaction buffer containing 50 mM TrisCl pH 8.6, 0.02% Triton X-100, 2 mM MgCl₂, 1 mM TCEP, 100 µM SAM for 3 hours at room temperature. Activity was detected by HTRF and MALDI-TOF.

Storage and Guarantee: Recombinant proteins in solution are temperature sensitive and must be stored at -80°C to prevent degradation. Avoid repeated freeze/thaw cycles and keep on ice when not in storage. This product is for research use only and is not for use in diagnostic procedures. This product is guaranteed for 6 months from date of arrival.



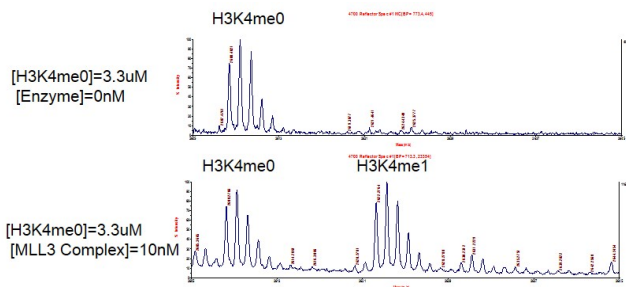
Recombinant KMT2C (MLL3) complex protein gel.

Recombinant KMT2C (MLL3) complex run on an SDS-PAGE gel and stained with Coomassie Blue.



Recombinant KMT2C (MLL3) complex activity assay.

3.3 μ M H3K4me0 (1-21aa) peptide was incubated with Recombinant KMT2C (MLL3) complex in reaction buffer for 3 hour at room temperature. Recombinant KMT2C (MLL3) complex was used in a HTRF assay to determine enzyme linearity. Methylated peptide (H3K4me2) was measured using H3K4me2-specific antibody.



Recombinant KMT2C (MLL3) complex activity assay.

3.3 μ M H3K4me0 peptide was incubated with 10 nM Recombinant KMT2C (MLL3) complex in reaction buffer for 3 hours at room temperature. The reaction product was detected by MALDI-TOF. Single 3.3 μ M H3K4me0 peptide was used as negative control.

Catalytic Ability: ~165 turnovers/ enzyme molecule