

Recombinant PRC2 EZH2(Y641N) complex

Catalog No: 31390, 31790

Expressed In: Baculovirus

Quantity: 20 µg

Concentration: 1.4 µg/µl

Source: Human

Buffer Contents: Full length recombinant PRC2 EZH2 (Y641N) Complex expressed in Sf9 at a concentration of 1.4 µg/µl in 25 mM HEPES-NaOH pH 7.5, 120 mM NaCl and 5% glycerol, 0.2 mg/ml 3X FLAG peptide.

Background: PRC2 (Polycomb Repressive Complex 2) is one of the two classes Polycomb-group, or PcG proteins (the other being PRC1) that are important epigenetic determinants of stem cell identity. They play an important role in long-term epigenetic silencing of genes during cell fate determination and differentiation. PRC2 functions as a repressor of chromatin. PRC2 is required to target recruitment to specific DNA sequences (termed Polycomb Response Elements or PREs) of genomic regions to be silenced. Once associated with chromatin, the PRC2 subunit EZH2 has histone methyltransferase activity that catalyzes the trimethylation of histone H3 at Lys27. H3K27me3 is well established as a hallmark of regions of repressed chromatin. Trimethylation of Lys27 leads to the recruitment of PRC1 through the binding of H3K27me3 by chromodomain-containing proteins in PRC1. PRC1 is responsible for long-term gene silencing after cellular differentiation.

Recombinant PRC2 EZH2(Y641N) Complex is a mutant version of our wild-type PRC2 Complex that contains a Tyro-Asn mutation at tyrosine 641 of the SET domain of EZH2. Somatic mutations of tyrosine 641, including Y641F, Y641N, Y641C, Y641S and Y641H, have been identified in patients with non-Hodgkin lymphoma and have been shown to alter substrate specificity and catalytic activity of EZH2 for histone H3 lysine 27 (H3K27) methylation states resulting in increased H3K27 trimethylation.

Protein Details: Recombinant PRC2 EZH2(Y641N) Complex that includes full length EZH2 with a Y641N mutation complexed with full length SUZ12, EED and RbAp46/48 (accession numbers NP_001190176.1, NP_056170, NP_003788.2, NP_002884.1, and NP_005601.1, respectively) was expressed in Sf9 and contains an N-terminal FLAG tag at the N-terminus of EZH2. The molecular weights of expressed EZH2 (Y641N), SUZ12, EED and RbAp46/48 are 87 kDa, 83 kDa, 50.2 kDa, 47.8 kDa and 47.7 kDa, respectively. The recombinant protein is >92% pure by SDS-PAGE.

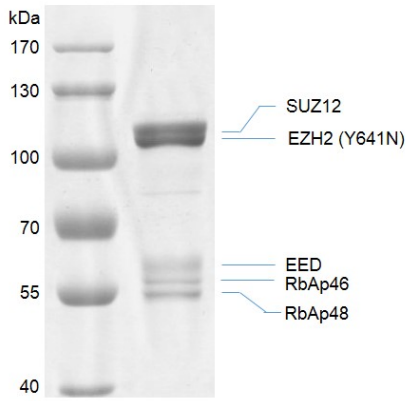
Application Notes: PRC2 EZH2(Y641N) Complex is suitable for use in the study of enzyme kinetics, inhibitor screening and selectivity profiling.

Specific Activity: H3K27me3 and H3K27me2 methyltransferase.

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 guaranteed for 6 months from date of receipt.

This product is for research use only and is not for use in diagnostic procedures.

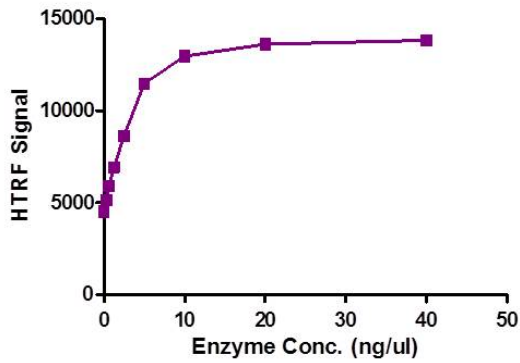
PRC2 EZH2 (Y641N) Complex



Recombinant PRC2 EZH2(Y641N) Complex gel.

PRC2 EZH2(Y641N) Complex was run on a 10% SDS-PAGE gel and stained with Coomassie Blue.

PRC2 EZH2 (Y641N) Titration



HTRF Assay for Recombinant PRC2 EZH2(Y641N) Complex activity.

1 μ M H3K27me2 peptide was incubated with PRC2 EZH2 (Y641N) Complex in reaction buffer including 50 mM Tris-HCL pH 8.6, 0.02% Triton X-100, 2 mM MgCl₂, 1 mM TCEP, 50 μ M SAM for 3 hours at room temperature. Anti-H3K27me3 antibody was used to detect reaction products

MALDI-TOF for PRC2 EZH2 (Y641N) activity. 3.3 μ M H3K27me2 peptide was incubated with 40 nM PRC2 EZH2 (Y641N) Complex in reaction buffer for 3 hours at room temperature. Activity was detected by MALDI-TOF. Catalytic Ability: 80 turnovers / enzyme molecule.

