## Recombinant Tet1 protein



Catalog No: 31363 Expressed In: *E. coli*  Quantity: 25 µg Concentration: 1 µg/µl Source: Mouse

**Buffer Contents:** 25 µg recombinant Tet1 protein expressed in *E. coli* and supplied in a buffer of 20 mM Tris 8.0, 300 mM NaCl, 2 mM DTT and 5% glycerol.

**Background: Ten-eleven Translocation Gene Protein 1 (Tet1)** is a DNA-binding protein that modulates DNA methylation and gene transcription via hydroxylation of 5-methylcytosine (5-mC). Tet1 is a cytosine oxygenase that catalyzes the conversion of the modified genomic base 5-mC into 5-hydroxymethylcytosine (5-hmC). Methylation at the C5 position of cytosine is an epigenetic modification of the mammalian genome that plays an important role in transcriptional regulation. Tet1 is responsible for initiating the enzymatic deamination process leading to cytosine demethylation. Tet1 preferentially binds to CpG-rich sequences at the promoters of both transcriptionally active and Polycomb-repressed genes and modulates gene silencing induced by cytosine methylation by controlling the levels of 5-mC and 5-hmC at gene promoters. Tet1 may have the dual function of repressing the expression of a subset of genes through recruitment of transcriptional repressors to promoters. Tet1 is important for stem cell maintenance and specification and is involved in the balance between pluripotency and lineage commitment.

**Protein Details:** Tet1 is a member of the cytosine oxygenase family of enzymes that convert 5-methylcytosine to 5-hydroxymethylcytosine. Amino acids 1367 to 2039 of mouse Tet1 (accession number ACY38291.1) were expressed in *E. coli* (MW = 95 kDa).

**Application Notes:** Recombinant Tet1 protein is suitable for use in cytosine oxygenase assays. A good starting point is 0.5 to 5  $\mu$ g of enzyme per assay incubated with 0.5  $\mu$ g of substrate in 50 mM HEPES, pH 8, 50  $\mu$ M Fe(NH4)<sub>2</sub>(SO4) <sub>2</sub>, 2 mM ascorbate, and 1 mM alpha-ketoglutarate for 3 hr at 37°C.

NOTE: Reaction buffers should made fresh.

## **References:**

This product was used in the following publications: *J. Neurosci.* (2016). 36(9):2769-81. PMID: 26937014.

**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.



## Tet1 activity assay.

Recombinant Tet1 activity measured using an antibody-based activity assay.

Top Panel:5-hmC DNA standards detected with an antibody recognizing 5-hydroxymethylcytosine. Amount of DNA (ng) is indicated above each spot.

Bottom Panel: Single-stranded DNA containing 5-mC was incubated with 5 µg of recombinant Tet1 enzyme (+Tet1), without Tet1 (-Tet), or unmethylated DNA not incubated with enzyme (DNA) and then spotted and incubated with an antibody recognizing 5-hmC to detect conversion of 5-mC into 5-hmC.