

Recombinant ALKBH3 protein

Catalog No: 81130, 81830

Lot No: 14318001

Expressed In: *E. coli*

Quantity: 100, 1000 µg

Concentration: 2 µg/µl

Source: Human

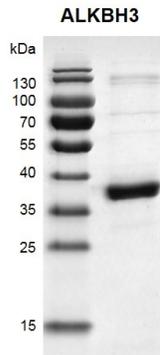
Buffer Contents: Recombinant ALKBH3 protein is supplied in 25 mM Tris pH 8.0, 300 mM NaCl, 10% glycerol, 0.5 mM TCEP.

Background: ALKBH3 (AlkB Homolog 3, Alpha-Ketoglutarate Dependent Dioxygenase) is a member of ALKBH protein family. It mediates demethylation of DNA and RNA containing 1-methyladenosine (m1A). It can repair alkylated DNA containing 1-methyladenosine (m1A) and 3-methylcytosine (m3C) by oxidative demethylation. ALKBH3 has a strong preference for single-stranded DNA. It is able to process alkylated m3C within double-stranded regions via its interaction with ASCC3, which promoting DNA unwinding to generate single-stranded substrate needed for ALKBH3. ALKBH3 also acts on RNA via demethylating N(1)-methyladenosine (m1A) RNA, an epigenetic internal modification of messenger RNAs (mRNAs) highly enriched within 5-untranslated regions (UTRs) and in the vicinity of start codons. The oxidative demethylation requires molecular oxygen, alpha-ketoglutarate and iron.

Protein Details: Full length ALKBH3 protein (accession number NP_631917.1) was expressed in *E.coli* cells with an N-terminal 6xHis tag. The molecular weight of ALKBH3 is 36.9 kDa.

Application Notes: Recombinant ALKBH3 protein is suitable for use in enzyme kinetics, inhibitor screening, and selectivity profiling.

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.



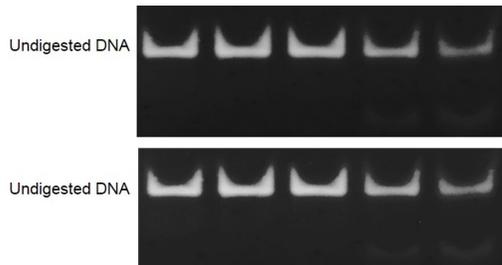
Recombinant ALKBH3 protein

12.5% SDS-PAGE Coomassie staining

MW: 36.9 kDa

Purity: >90%

| | | | | | | |
|--------|---|-----|---|---|---|----|
| DNA | 1 | 1 | 1 | 1 | 1 | µg |
| ALKBH3 | 0 | 0.5 | 1 | 2 | 4 | µg |



ALKBH3 dioxygenase activity

1 µg of ssDNA oligos (5'-AAAGCAG(1mA)

ATTCGAAAAGCGAAA-3') was incubated with varying concentrations of ALKBH3 in a reaction system including 50 mM HEPES-NaOH pH 8.0, 50 µM Fe(NH₄)₂(SO₄)₂, 1 mM 2-oxoglutarate, 2 mM ascorbate and 1 mM TCEP for 30 min at 37°C. Then ssDNA oligos were annealed with equimolar of non-methylated complement strand followed by 1 µg EcoRI digestion for 45 min at 37°C.

% reaction products were run on a 20% Native PAGE gel and stained by ethidium bromide.