

Recombinant Mononucleosomes H3K4me2 (EPL) - biotin

Catalog No: 81074

Lot No: 03318001

Expressed In: *E. coli*

Quantity: 20 µg

Concentration: 0.56 µg/µl

Source: Human

Buffer Contents: Recombinant Mononucleosomes H3K4me2 (EPL) - biotin (20 µg protein + 20 µg DNA) are supplied in 10 mM Tris-HCl pH 8.0, 1 mM EDTA, 2 mM DTT and 20% glycerol.

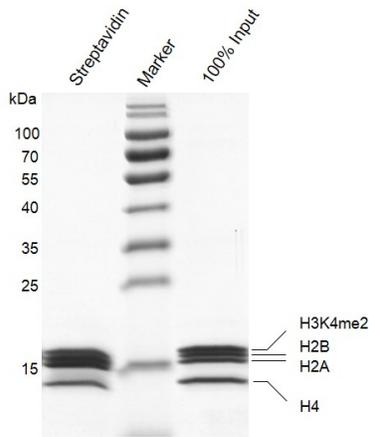
Background: *In vivo*, histones are wrapped around by DNA in chromatin. Therefore, nucleosomes are more physiologically relevant substrates than histones and histone-derived peptides for *in vitro* studies. More importantly, some histone methyltransferases are significantly more active, as well as specific, when using nucleosomal substrates in HMT assays, such as DOT1L and NSD family enzymes. Nucleosomes are also widely used in histone methyltransferase screening assays to identify small molecular inhibitors for drug discovery.

Protein Details: Recombinant Mononucleosomes H3K4me2 (EPL) - biotin consist of a 167 bp of 601 DNA with 5'-biotin tag and two molecules each of histones H2A that includes amino acids 1-130 (end) (accession number NP_003503.1), H2B that includes amino acids 1-126 (end) (accession number NP_003509.1), H3.2 that includes amino acids 1-136 (end) (accession number NP_066403.2) with dimethylation at lysine 4, and H4 that includes amino acids 1-103 (end) (accession number NP_003539.1). All of these histones were expressed in *E. coli* cells. The molecular weight of histone octamer is ~108 kDa.

H3K4me2 (Histone H3 dimethyl Lys4) protein is generated using expressed protein ligation (EPL) technology. Truncated human Histone H3.2 is produced in *E. coli* and purified using FPLC. The purified protein is subsequently ligated to a N-terminal histone tail peptide containing dimethyl lysine 4 via a native peptide bond.

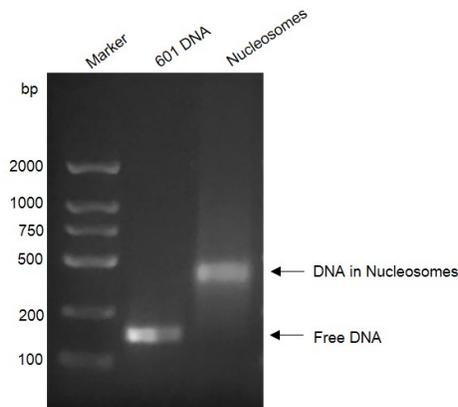
Application Notes: Recombinant Mononucleosomes H3K4me2 (EPL) - biotin are suitable for use in the study of enzyme kinetics, inhibitor screening, and selectivity profiling.

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.



Streptavidin pull-down for Recombinant Mononucleosomes H3K4me2 (EPL) - biotinylated.

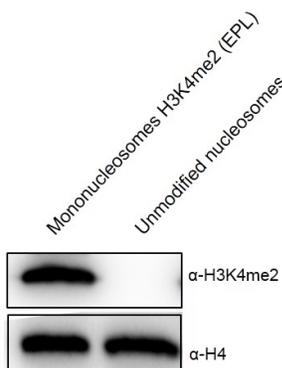
Recombinant Mononucleosomes H3K4me2 (EPL) - biotin were pulled down by streptavidin beads. Input nucleosomes (Lane 1) and the nucleosomes (Lane 2) pulled down by streptavidin were run on a 12.5% SDS-PAGE gel and stained with Comassie blue. The SDS-PAGE gel result shows that almost all of biotinylated nucleosomes H3K4me2 are pulled down by streptavidin beads. The purity of mononucleosomes H3K4me2 (EPL) - biotin is $\geq 95\%$.



Recombinant Mononucleosomes H3K4me2 (EPL) - biotin DNA gel

Mononucleosomes H3K4me2 (EPL) -biotin were run on a 2% agarose gel and stained with ethidium bromide. Lane 1: DNA marker. Lane 2: 601 DNA. Lane 3: Intact nucleosome. Intact nucleosomes migrate much higher than free 601 DNA.

The agarose gel result shows almost all of 601 DNA wraps histone octamers to form nucleosomes.



Western Blot analysis for Recombinant Mono-nucleosomes H3K4me2 (EPL) – biotin

Recombinant Mononucleosomes H3K4me2 (EPL) - biotin (Lane 1) and unmodified nucleosomes (Lane 2) were detected with anti-H3K4me2 antibody (Cat# 39679) and anti-H4 antibody (Cat# 61300), respectively. H3 was detected as loading control.

Only Recombinant Mononucleosomes H3K4me2 (EPL) - biotin can be detected by anti-H3K4me2 antibody.