

## Recombinant Mononucleosomes H3K9me3 (EPL) - biotinylated

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**Catalog No:** 31555, 31955

**Lot No:** 17717002

**Expressed In:** *E. coli*

**Quantity:** 20, 1000 µg

**Concentration:** 0.35 µg/µl

**Source:** Human

**Buffer Contents:** Recombinant Mononucleosomes H3K9me3 (EPL) - biotinylated (20 µg protein + 20 µg DNA) is supplied at a protein concentration of 0.35 µg/µl in 10 mM Tris-HCl, pH 8.0, 1 mM EDTA, 2 mM DTT and 20% glycerol.

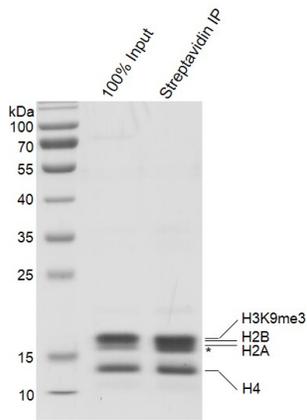
**Background:** *In vivo*, histones are wrapped around by DNA in chromatin. Therefore, nucleosomes are more physiologically relevant substrates than histones and histone-derived peptides for *in vitro* studies. More importantly, some histone methyltransferases are significantly more active, as well as specific, when using nucleosomal substrates in Histone Methyltransferase (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 H3K9me3 (EPL) - biotinylated 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 trimethylation at lysine 9, 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. The recombinant protein is ≥95% by SDS-PAGE gel. H3K9me3 (Histone H3 trimethyl Lys9) 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 trimethyl lysine 9 via a native peptide bond.

**Application Notes:** Recombinant Nucleosome H3K9me3 (EPL) is suitable for use in the study of enzyme kinetics, inhibitor screening, and selectivity profiling.

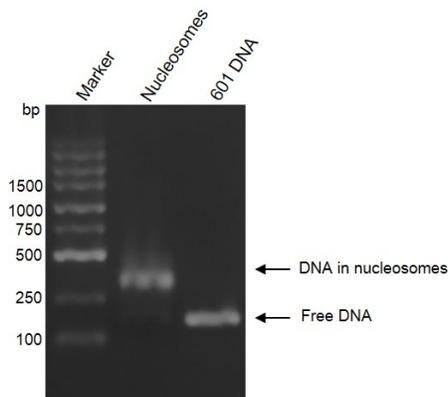
**Storage and Guarantee:** Recombinant proteins in solution are temperature sensitive and must be stored at -80°C to prevent degradation. Avoid repeated freeze/thaw cycles and keep on ice when not in storage. This product is guaranteed for 6 months from date of receipt.

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



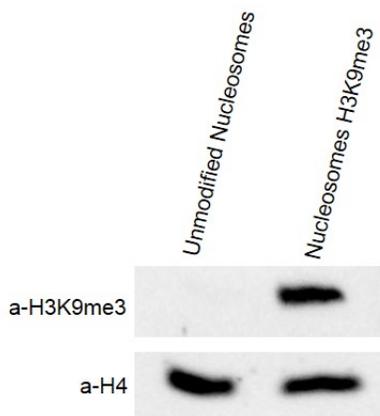
### Streptavidin pull-down for Recombinant Mononucleosomes H3K9me3 (EPL) - biotin.

Recombinant Mononucleosomes H3K9me3 (EPL) - biotin were pulled down by streptavidin beads. Input nucleosomes (Lane 2) and the nucleosomes (Lane 3) pulled down by streptavidin were run on a 13% SDS-PAGE gel and stained with Commassie blue. The SDS-PAGE gel result shows that almost all of biotinylated mononucleosomes H3K9me3 are pulled down by streptavidin beads. \* indicates streptavidin.



### Recombinant Mononucleosomes H3K9me3 (EPL) - biotin, DNA gel.

Recombinant Mononucleosomes H3K9me3 (EPL) - biotinylated were run on a 2% agarose gel and stained with ethidium bromide. Lane 1: DNA marker. Lane 2: Intact mononucleosomes H3K9me3. Lane 3: 601 DNA. Intact mononucleosomes H3K9me3 migrated much higher than free 601 DNA. The agarose gel result shows almost all of 601 DNA wrap histone octamers to form nucleosomes.



### Western Blot analysis for Recombinant Mononucleosomes H3K9me3 (EPL) - biotin.

Unmodified nucleosomes (Lane 1) and Recombinant Mononucleosomes H3K9me3 (EPL) - biotinylated (Lane 2) were detected with anti-H3K9me3 antibody and anti-H4 antibody, respectively. H4 was detected as loading control. Only Recombinant Mononucleosomes H3K9me3 (EPL) - biotinylated can be detected by anti-H3K9me3 antibody.