H3K9me3 Recombinant Nucleosome, Biotinylated

Catalog No	16-0315	**Species**	Human
Lot No	24173008-01	**Source**	*E. coli* & synthetic DNA
Pack Size	50 µg	**Tag**	Biotinylated
Concentration	4.6 µM	**MW**	199,732 Da

DESCRIPTION

H3K9me3 (histone H3 lysine 9 trimethylation) Recombinant Nucleosome, Biotinylated consists of 147 base pairs of DNA wrapped around an octamer core of histone proteins (two each of H2A, H2B, H3.2, and H4) to form a nucleosome, the basic repeating unit of chromatin. The 147 bp 601 sequence, identified by Lowary and Widom [1], has high affinity for histone octamers and is useful for nucleosome assembly. H3K9me3 nucleosome contains trimethylated lysine at position 9 and a Cys to Ala substitution at position 110 on histone H3.2. The DNA contains a 5' biotin-TEG group.

TECHNICAL INFORMATION

Storage	Stable for six months at -80°C from date of receipt. For best results, aliquot and avoid freeze/thaws.
Formulation	0.92 mg/mL mononucleosome in 54.3 µL 10 mM Tris HCl pH 7.5, 25 mM NaCl, 1 mM EDTA, 2 mM DTT, 20% glycerol (27.2 µg protein, 50 µg DNA + protein).

APPLICATION NOTES

H3K9me3 nucleosome is highly purified and suitable for a variety of applications, including use as a substrate in enzyme assays, high-throughput screening and inhibitor testing, chromatin binding studies, protein-protein interaction assays, structural studies, and in effector protein binding experiments. For a corresponding unmodified control, we recommend EpiCypher 16-0006.

GENE & PROTEIN INFORMATION

UniProt ID	H2A - P04908 (alt. names: H2A type 1-B/E, H2A.2, H2A/a, H2A/m) H2B - O60814 (alt. names: H2B K, HIRA-interacting protein 1) H3.2 - Q71DI3 H4 - P62805

REFERENCES

[1] Lowary & Widom *J. Mol. Biol.* (1998). PMID: 9514715

VALIDATION DATA

FIGURE 1 Western blot data. Western Analysis of H3K9me3 nucleosome. **Top Panel:** Unmodified (EpiCypher 16-0006; Lane 1) and H3K9me3 (Lane 2) nucleosomes were probed with an anti-H3K9me3 antibody and analyzed via enhanced chemiluminescence (ECL) readout. Only the H3K9me3 sample produced a detectable signal. **Bottom Panel:** Detail from Coomassie stained gel showing unmodified (Lane 1) and H3K9me3 (Lane 2) nucleosomes.

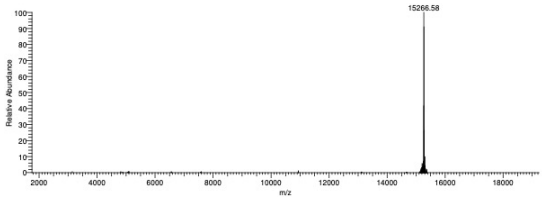

FIGURE 2 Mass spec data. Synthetic H3K9me3 histone analyzed by high resolution mass spectrometry. Expected mass = 15,267.8 Da. Determined mass = 15,266.58 Da.

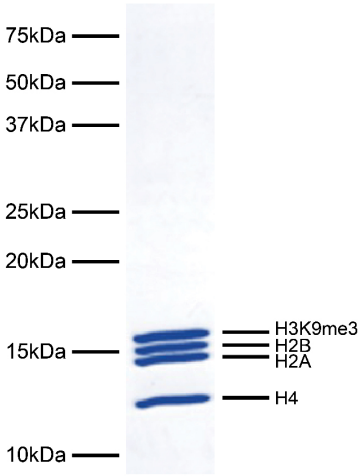

FIGURE 3 Protein gel data. Coomassie stained SDS-PAGE gel of proteins in H3K9me3 nucleosome (1 µg) demonstrates the purity of histones in the preparation. Sizes of molecular weight markers and positions of the core histones (H2A, H2B, H3K9me3, and H4) are indicated.

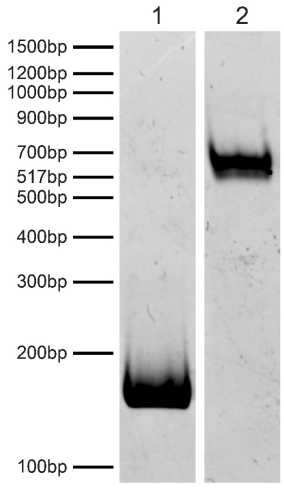

FIGURE 4 DNA gel data. H3K9me3 nucleosome resolved via native PAGE and stained with ethidium bromide to visualize DNA. Both lanes are from the same gel. **Lane 1:** Free DNA (EpiCypher 18-0005; 100 ng). **Lane 2:** Intact H3K9me3 nucleosomes (400 ng).