H3.3S31phos,K36me3 Recombinant Nucleosome, Biotinylated

Catalog No	16-0407	**Species**	Human
Lot No	24121001-02	**Source**	*E. coli* & synthetic DNA
Pack Size	50 µg	**Tag**	Biotinylated
Concentration	4.5 µM	**MW**	199,836 Da

DESCRIPTION

Histone phosphorylation is a post-translational modification (PTM) wherein a phosphate group is added to a histone protein, predominantly occurring on serine, threonine, and tyrosine residues. In combination with other PTMs, histone phosphorylation constitutes the "histone code," acting as a language read by proteins to regulate chromatin structure and gene expression. Histone phosphorylation is involved in chromatin remodeling and compaction associated with diverse cellular processes, including DNA damage repair, transcription regulation, cell division, and apoptosis [1]. Histone phosphorylation is also observed on non-canonical histones, particularly H3.3, where it plays roles in transcriptional regulation. Recombinant mononucleosomes containing phosphorylated histones can be used to study the biological functions of histone phosphorylation.

H3.3S31phos,K36me3 (histone H3.3 serine 31 phosphorylation, lysine 36 trimethylation) Recombinant Nucleosome, Biotinylated consists of 147 base pairs of DNA wrapped around an octamer of core histone proteins (two each of H2A, H2B, H3.3, and H4) to form a nucleosome, the basic repeating unit of chromatin. The 147 bp 601 sequence, identified by Lowary and Widom [2], has high affinity for histone octamers and is useful for nucleosome assembly. The DNA contains a 5' biotin-TEG group. The variant H3.3 contains a serine at position 31, one of several discrete amino acid differences compared to canonical H3.1. H3.3S31phos,K36me3 contains phosphorylated serine at position 31 and trimethylated lysine at position 36 on histone H3.3. H3.3S31phos,K36me3 modulates stimulation-responsive gene expression in activated macrophages via engagement of the histone methyltransferase SETD2 and ejection of the transcriptional co-repressor ZMYND11 [3].

TECHNICAL INFORMATION

Storage	Stable for six months at -80°C from date of receipt. For best results, aliquot and avoid freeze/thaws.
Formulation	0.89 mg/mL mononucleosome in 56.2 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

H3.3S31phos,K36me3 mononucleosome 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) H3.3 - P84243 H4 - P62805

VALIDATION DATA

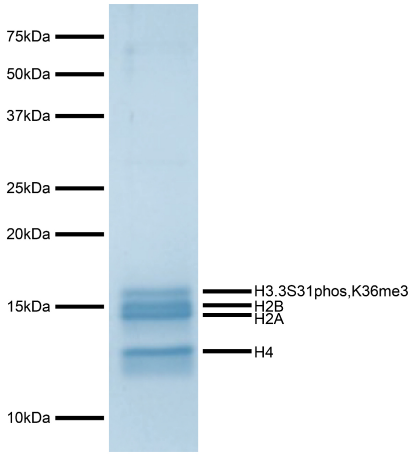

FIGURE 1 Protein gel data. Coomassie stained PAGE gel of proteins in H3.3S31phos, K36me3 nucleosome (1 µg) demonstrates the purity of histones in the preparation. Sizes of molecular weight markers and positions of the core histones (H2A, H2B, H3.3S31phos,K36me3, and H4) are indicated.

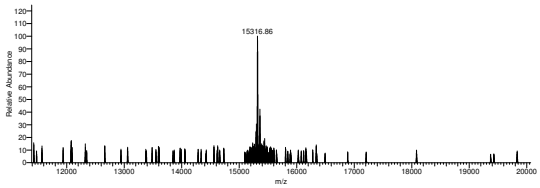

FIGURE 2 Mass spec data. Synthetic H3.3S31phos,K36me3 histone analyzed by high resolution mass spectrometry. Expected mass = 15,319.7 Da. Determined mass = 15,316.86 Da.

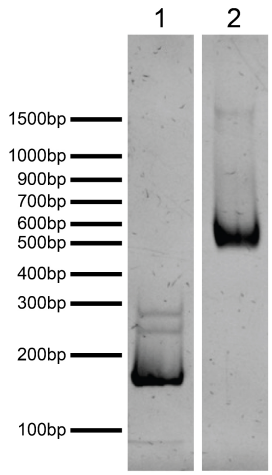

FIGURE 3 DNA gel data. H3.3S31phos,K36me3 nucleosome resolved via native PAGE and stained with ethidium bromide to visualize DNA. Both lanes are from the same gel. **Lane 1:** Free DNA (EpiCypher 18-0005; 100 ng). Free DNA is over 95% pure by densitometry. **Lane 2:** Intact H3.3S31phos,K36me3 nucleosomes (400 ng).

REFERENCES

[1] Rossetto et al. *Epigenetics* (2012). PMID: 22948226
[2] Lowary & Widom *J. Mol. Biol.* (1998). PMID: 9514715
[3] Armache et al. *Nature* (2020). PMID: 32699416