

Mononucleosomes (H3.3K27M), Recombinant Human, Biotinylated

Catalog No	16-1323	Species	Human
Lot No	22206003-04	Source	<i>E. coli</i> & synthetic DNA
Pack Size	50 µg	Tag	Biotinylated
Concentration	5.0 µM	MW	199,712.94 Da

DESCRIPTION

Oncogenic Nucleosomes (oncoNucs™) are recombinant nucleosomes assembled using histones with cancer-associated mutations, providing physiologically relevant substrates for cancer research. OncoNucs consist of 147 base pairs of DNA wrapped around an octamer core of histone proteins (two each of H2A, H2B, H3.3 and H4). The 147 bp 601 sequence, identified by Lowary and Widom [1], has high affinity for histone octamers and is useful for nucleosome assembly. H3.3K27M mononucleosome contains a methionine at position 27 on histone H3.3, which is a dominant negative inhibitor of the EZH2 methyltransferase of the PRC2 complex and associated with pediatric brain cancer (DIPG). The DNA in this nucleosome 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	1.0 mg/mL mononucleosome in 50.1 µL 10 mM Tris HCl pH 7.5, 25 mM NaCl, 1 mM EDTA, 2 mM DTT, 20% glycerol (27.5 µg protein, 50.0 µg DNA+ protein)

APPLICATION NOTES

Mononucleosomes (H3.3K27M) are 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.

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
-------------------	--

REFERENCES

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



FIGURE 1: Western blot data. Western analysis of H3.3K27M Mononucleosomes. **Top Panel:** Unmodified H3.3 (EpiCypher 15-0312, Lane 1) and H3.3K27M nucleosomes (Lane 2) were probed with an anti-H3K27M antibody and analyzed via ECL readout. Only the H3.3K27M sample produced a detectable signal. **Bottom Panel:** Detail from Coomassie stained gel showing unmodified H3.3 (Lane 1) and H3.3K27M nucleosomes (Lane 2).

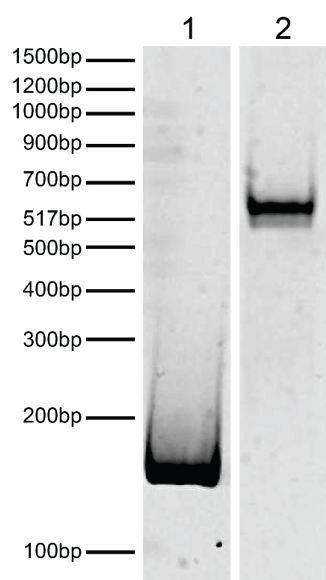


FIGURE 2: DNA gel data. H3.3K27M mononucleosomes resolved via native PAGE and stained with ethidium bromide to visualize DNA. **Lane 1:** Free DNA (EpiCypher 18-0005; 100ng) **Lane 2:** Intact H3.3K27M mononucleosomes (400 ng).

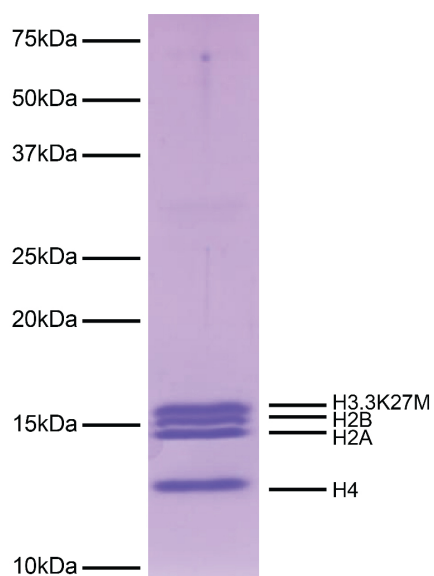


FIGURE 3: Protein gel data. Coomassie stained SDS-PAGE gel of proteins in H3.3K27M mononucleosomes (1 µg) demonstrates the purity of histones in the preparation. Sizes of molecular weight markers and positions of the core histones (H2A, H2B, H3.3K27M and H4) are indicated.