H2AK119ub1 Recombinant Nucleosome, Biotinylated

Catalog No	16-0395	**Species**	Human
Lot No	24284005-02	**Source**	*E. coli* & synthetic DNA
Pack Size	25 µg	**Tag**	Biotinylated
Concentration	6.3 µM	**MW**	216,834 Da

DESCRIPTION

Histone ubiquitination is a post-translational modification (PTM) wherein ubiquitin is added to a lysine residue of a histone protein. In combination with other PTMs, histone ubiquitination constitutes the "histone code," acting as a language read by proteins to regulate chromatin structure and gene expression. Ubiquitin is added through the sequential actions of E1 activating, E2 conjugating, and E3 ligating enzymes and is removed by deubiquitinating enzymes (DUBs). Histone ubiquitination plays an integral role in DNA damage response and has been implicated in transcriptional regulation and DNA replication [1]. Recombinant nucleosomes containing ubiquitinated histones are useful to study the biological functions of histone ubiquitination.

H2AK119ub1 (histone H2A lysine 119 ubiquitination) Recombinant Nucleosome, Biotinylated consists of 147 base pairs of 601 sequence DNA [2] wrapped around an octamer of core histone proteins (two each of H2A, H2B, H3.1, and H4) to form a nucleosome, the basic repeating unit of chromatin. The DNA contains a 5' biotin-TEG group. H2AK119ub1 nucleosome contains ubiquitinated lysine at position 119 on histone H2A. H2AK119 is ubiquitinated by E3 ligase RING1A/B, subunits of the polycomb repressive complex 1 (PRC1), in vivo. H2AK119ub1 is an abundant histone mark that plays a role in transcriptional regulation [1].

TECHNICAL INFORMATION

Storage	Stable for six months at -80°C from date of receipt. For best results, aliquot and avoid freeze/thaws.
Formulation	1.37 mg/mL mononucleosome in 18.25 µL 10 mM Tris HCl pH 7.5, 25 mM NaCl, 1 mM EDTA, 2 mM DTT, 20% glycerol (14.5 µg protein, 25 µg DNA + protein).

APPLICATION NOTES

H2AK119ub1 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) H3.1 - P68431 (alt. names: H3, H3/a, H3/b, H3/c, H3/d) H4 - P62805

REFERENCES

[1] Mattiroli & Penengo *Trends Genet.* (2021). PMID: 33485674
[2] Lowary & Widom *J. Mol. Biol.* (1998). PMID: 9514715

VALIDATION DATA

FIGURE 1 Western blot data. Western Analysis of H2AK119ub1 nucleosome. **Top Panel:** Unmodified (EpiCypher 16-0006; Lane 1) and H2AK119ub1 nucleosomes (Lane 2) were probed with an anti-H2AK119ub1 antibody and analyzed via enhanced chemiluminescence (ECL) readout. Only the H2AK119ub1 sample produced a detectable signal. **Bottom Panel:** Detail from Coomassie stained gel showing unmodified (Lane 1) and H2AK119ub1 nucleosomes (Lane 2).

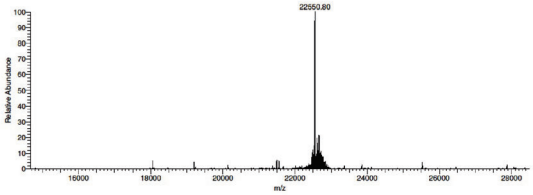

FIGURE 2 Mass spec data. Synthetic H2AK119ub1 histone analyzed by high resolution mass spectrometry. Expected mass = 22,550.3 Da. Determined mass = 22,550.80 Da.

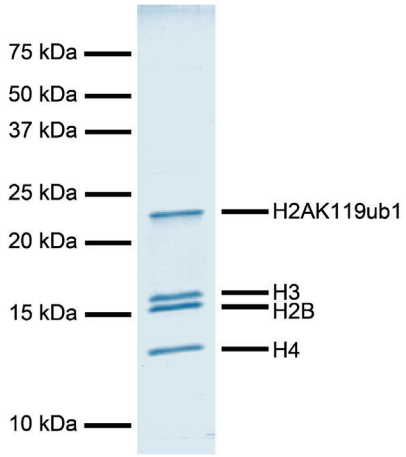

FIGURE 3 Protein gel data. Coomassie stained PAGE gel of proteins in H2AK119ub1 nucleosome (1 µg) demonstrates the purity of histones in the preparation. Sizes of molecular weight markers and positions of the core histones (H2AK119ub1, H3, H2B, and H4) are indicated.

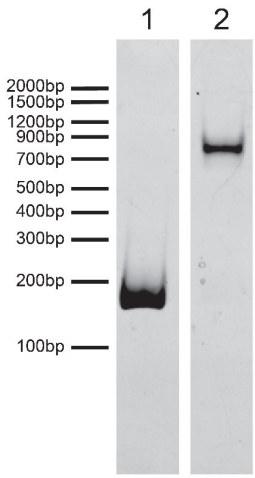

FIGURE 4 DNA gel data. H2AK119ub1 nucleosome resolved via native PAGE and stained with ethidium bromide to visualize DNA. **Lane 1:** Free DNA (EpiCypher 18-0005; 100 ng). **Lane 2:** Intact H2AK119ub1 nucleosomes (400 ng).