

Nucleosome, Recombinant Human, H3K14ub1 dNuc, Biotinylated

Catalog No	16-0398	Species	Human
Lot No	22193002-05	Source	<i>E. coli</i> & synthetic DNA
Pack Size	25 µg	Tag	Biotinylated
Concentration	5.1 µM	MW	216,857.3 Da

DESCRIPTION

H3K14ub1 designer mononucleosomes (dNucs) consist of 147 base pairs of DNA wrapped around an octamer core of histone proteins (two each of H2A, H2B, H3.1 and H4) to form a nucleosome, the basic repeating unit of chromatin. H3K14ub1 has been shown to promote the activity of the methyltransferase Clr4, which methylates H3K9, a known repressive mark [1]. The 147 bp 601 sequence, identified by Lowary & Widom [2], has high affinity for histone octamers and is useful for nucleosome assembly. The DNA in this nucleosome contains a 5' biotin-TEG group. H3K14ub1 has a Cys to Ala substitution at position 110.

TECHNICAL INFORMATION

Storage	Stable for six months at -80°C from date of receipt. For best results, aliquot and avoid freeze/thaws.
Formulation	1.115 mg/mL mononucleosome in 22.6 µL 10 mM Tris HCl pH 7.5, 25 mM NaCl, 1 mM EDTA, 2 mM DTT, 20% glycerol. (14.4 µg protein, 25 µg DNA + protein).

APPLICATION NOTES

H3K14ub1 dNuc 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.

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
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REFERENCES

- [1] Oya et al. *EMBO Rep.* (2019). PMID: 31468675
- [2] Lowary & Widom *J. Mol. Biol.* (1998). PMID: 9514715

VALIDATION DATA

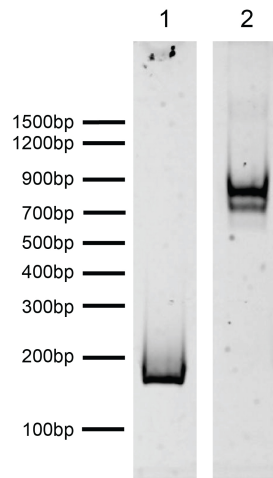


FIGURE 1: DNA gel data. H3K14ub1 dNuc resolved via native PAGE gel and stained with ethidium bromide to visualize DNA. **Lane 1:** Free DNA (EpiCypher 18-0005; 100 ng). **Lane 2:** Intact H3K14ub1 nucleosomes (400 ng). **NOTE:** The split band for the H3K14ub1 is a gel running artifact of the ubiquitin impacting the resolution of the nucleosome.

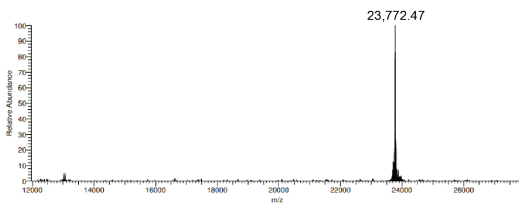


FIGURE 2: Mass spec data. Semi-synthetic H3K14ub1 histone analyzed by high resolution mass spectrometry. Expected mass = 23,771.60 Da. Determined mass = 23,772.47 Da.

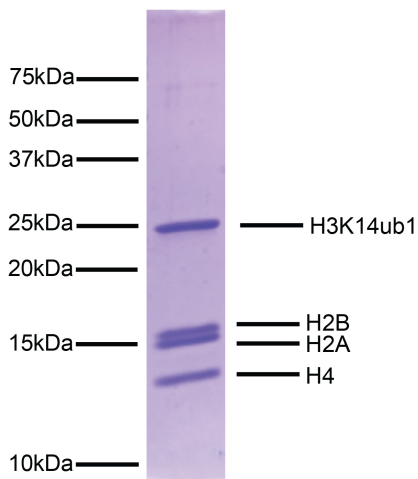


FIGURE 3: Protein gel data. Coomassie stained SDS-PAGE gel of proteins in H3K14ub1 dNuc (1 μ g) demonstrates the purity of histones in the preparation. Sizes of molecular weight markers and positions of the core histones (H2A, H2B, H3K14ub1 and H4) are indicated.