

Nucleosome, Recombinant Human, H3Cit2,8,17 dNuc, Non-Biotinylated

Catalog No	16-1362	Species	Human
Lot No	22346005-02	Source	E. coli & synthetic DNA
Pack Size	50 μg	Tag	None
Concentration	5.0 μΜ	MW	199,288.6 Da

DESCRIPTION

Recombinant mononucleosomes (H3Cit2,8,17) consist 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. H3Cit2,8,17 dNuc contains citrulline (instead of arginine) at positions 2, 8, and 17 on histone H3.2 and a Cys to Ala substitution at position 110.

TECHNICAL INFORMATION

Storage Formulation Stable for six months at -80°C from date of receipt. For best results, aliquot and avoid freeze/thaws. 1.0 mg/mL mononucleosome in 50.2 μ L 10 mM Tris HCl pH 7.5, 25 mM NaCl, 1 mM EDTA, 2 mM DTT, 20% glycerol. (27.3 μ g protein, 50 μ g DNA + protein)

APPLICATION NOTES

H3Cit2,8,17 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.

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



FIGURE 1 Western blot data. Western Analysis of H3Cit2,8,17 dNuc. Top Panel: Unmodified (EpiCypher 16-0009; Lane 1) and H3Cit2,8,17 nucleosomes (Lane 2) were probed with an anti-citrulline antibody and analyzed via ECL readout. Only the H3Cit2,8,17 sample produced a detectable signal. Bottom Panel: Detail from Coomassie stained gel showing unmodified (Lane 1) and H3Cit2,8,17 nucleosomes (Lane 2).

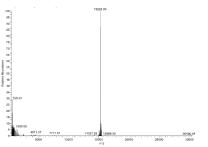


FIGURE 2 Mass spec data. Synthetic H3Cit2,8,17 histone analyzed by high resolution mass spectrometry. Expected mass = 15,227.8 Da. Determined mass = 15,228.00 Da.

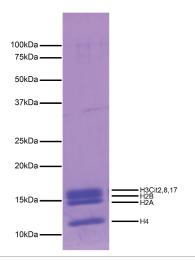


FIGURE 3 Protein gel data. Coomassie stained SDS-PAGE gel of proteins in H3Cit2,8,17 dNuc (1 μ g) demonstrates the purity of histones in the preparation. Sizes of molecular weight markers and positions of the core histones (H2A, H2B, H3Cit2,8,17, and H4) are indicated.

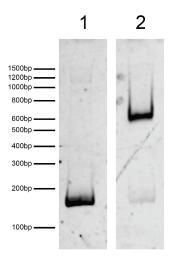


FIGURE 4: DNA gel data. H3Cit2,8,17 dNuc resolved via native PAGE gel and stained with ethidium bromide to visualize DNA. Lane 1: Free DNA (EpiCypher 18-0006; 100 ng). Lane 2: Intact H3Cit2,8,17 nucleosomes (400 ng).