Mononucleosomes (H3.3K4M), Recombinant Human Biotinylated

Catalog No.16-0349Lot No.21111003-01Pack Size50 μg

Product Description:

Mononucleosomes assembled from recombinant human histones expressed in E. coli (two each of histones H2A, H2B, H3.3 and H4; accession numbers: H2A-P04908, H2B-O60814, H3.3-P84243, H4-P62805) wrapped by 147 base pairs of 601 positioning sequence DNA (Lowary and Widom, 1998). H3.3 is a histone variant, found in regions of high chromatin turnover outside of S-phase (e.g. at actively transcribed genes). Recombinant human histone H3.3 (H3F3A, H3.3A, H3F3) contains a lysine-tomethionine (K-to-M) substitution at position four, a known cancer-associated oncohistone mutation that affects chromatin regulatory mechanisms (Nacev et al., 2018). The 601 sequence DNA contains a 5' biotin-TEG group, enabling affinity binding applications.

Formulation:

H3.3K4M mononucleosomes (27.3 μ g protein weight, 50 μ g DNA+protein) in 58.0 μ L 10 mM Tris HCl, pH 7.5, 25 mM NaCl, 1 mM EDTA, 2 mM DTT, 20% glycerol. Molarity = 4.3 μ M. MW = 200,543.

Storage and Stability:

Stable for six months at -80°C from date of receipt. For best results, aliquot and avoid multiple freeze/thaws.

Application Notes:

H3.3K4M mononucleosomes are highly purified and are suitable for a variety of applications, including use as a substrate in enzymatic assays or for effector protein binding experiments.

References:

Lowary PT and J Widom (1998) J Mol Biol 276: 19-42. Nacev BA et al. (2019) Nature 7749: 473-478.





Western Blot Data: Western analysis of H3.3K4M mononucleosomes. **Top Panel:** Unmodified H3.3 (Lane 1) and H3.3K4M - containing nucleosomes (Lane 2) were probed with an anti-H3K4M antibody. Only the H3.3K4M sample produced a detectable signal. **Bottom Panel:** Detail from Coomassie stained gel of Western blot.

DNA Gel Data: H3.3K4M mononucleosomes resolved by native PAGE and stained with ethidium bromide to visualize DNA. Lane 1: Free DNA extracted from nucleosomes (100 ng). Lane 2: Intact nucleosomes (400 ng).



Protein Gel Data: Coomassie stained PAGE gel of proteins in H3.3K4M mononucleosomes (1 μg) to demonstrate the purity of the histones in the preparation. Sizes of molecular weight markers and positions of the core histones (H2A, H2B, H3.3K4M and H4) are indicated.

This product is for in vitro research use only and is not intended for use in humans or animals.