

Histone H4

Human, Recombinant



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www.neb.com



M2504S 004140816081

M2504S



100 µg **1.0 mg/ml** **Lot: 0041408**
RECOMBINANT **Store at -20°C** **Exp: 8/16**

Description: Histone H3 combines with Histone H4 to form the H3/H4 tetramer. Two H2A/H2B heterodimers interact with an H3/H4 tetramer to form the histone octamer (1,2). Histone H4 is also modified by various enzymes and can act as a substrate for them. These modifications have been shown to be important in gene regulation.

Source: An *E. coli* strain that carries a plasmid encoding the cloned human histone H4 gene, HIST2H4. (Genbank accession number: AF525682)

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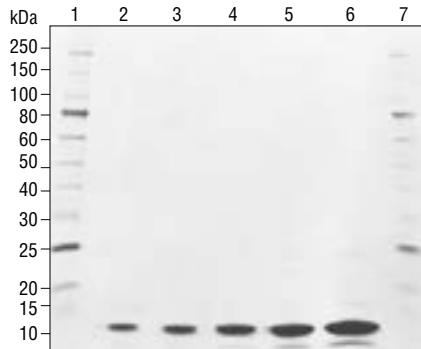
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Supplied in: 20 mM Sodium Phosphate (pH 7.0), 300 mM NaCl and 1 mM EDTA.

Note: The protein concentration (1 mg/ml, 89 µM) is calculated using the molar extinction coefficient for Histone H4 (5120) and its absorbance at 280 nm (3,4). 1.0 A₂₈₀ units = 2.2 mg/ml

Synonyms for HIST2H4 gene: H4/N, H4F2, H4FN

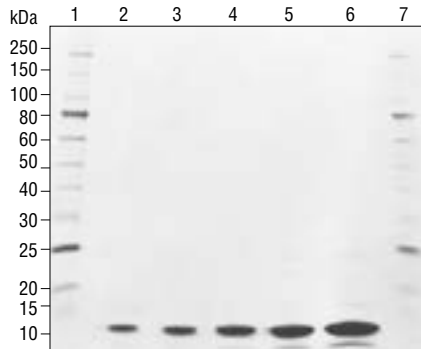


SDS-PAGE analysis of Histone H4 Human, Recombinant. Lane 1 & 7: NEB Protein Ladder (NEB #P7703), Lane 2 thru 6: 0.5–10.0 µg Histone H4 Human, Recombinant. (Please see Quality Control section for more information)

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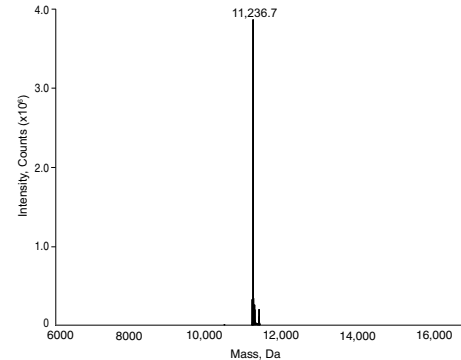


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Quality Control Assays:

SDS-PAGE: 0.5, 1.0, 2.0, 5.0, 10.0 µg of Histone H4 Human, Recombinant were loaded on a 10–20% Tris-Glycine SDS-PAGE gel and stained with Coomassie Blue. The calculated molecular weight is 11236.15 Da. Its apparent molecular weight on 10–20% Tris-Glycine SDS-PAGE gel is ~11 kDa (see figure 1).

Mass Spectrometry: The mass of purified Histone H4 Human, Recombinant is 11,236.7 Da as determined by ESI-TOF MS (Electrospray

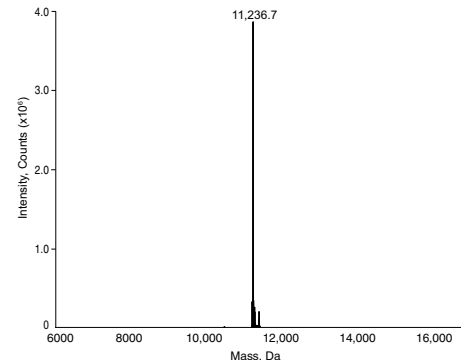


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ESI-TOF Analysis of Histone H4 Human, Recombinant.

Ionization-Time of Flight Mass Spectrometry). The average mass calculated from primary sequence is 11236.15 Da. This confirms the protein identity as well as the absence of any modifications of the histone.

N-terminal Protein Sequencing: Protein identity was confirmed using Edman Degradation to sequence the intact protein.

Enzyme Modification: After incubation of a 25 µl reaction for 10 minutes at 37°C, 1 unit of PRMT1 methyltransferase (NEB #M0234) transfers 2 pmols of methyl group to Histone H4 Human, Recombinant.

Protease Assay: After incubation of 10 µg of Histone H4 Human, Recombinant with a standard mixture of proteases for 2 hours at 37°C, no proteolytic activity could be detected by SDS-PAGE.

Exonuclease Assay: Incubation of a 50 µl reaction containing 10 µg of Histone H4 Human, Recombinant with 1 µg of a mixture of single and double-stranded [³H] *E. coli* DNA (200,000 cpm/µg) for 4 hours at 37°C released < 0.1% of the total radioactivity.

(See other side)

CERTIFICATE OF ANALYSIS

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Endonuclease Assay: Incubation of a 50 µl reaction containing 10 µg of Histone H4 Human, Recombinant with 1 µg of ϕX174 RF I (suprecoiled) plasmid DNA for 4 hours at 37°C resulted in < 5.0% conversion to RF II form (nicked circle) as determined by agarose gel electrophoresis.

Protein Sequence:SGRGKGGKGLGKGGAKRH
RKVLRDNIQGITKPAIRRLARRGGVKRISGLIYEE
TRGVLVFLENVIRDAVITYTEHAKRKTVTAMDVV
YALKRQGRTLYGFGG (Genbank accession
number: AAM83108)

References:

1. Kornberg, R.D. (1977) *Annu. Rev. Biochem.* 46, 931–954.
2. van Holde, K.E. (1989) *Chromatin*, 1–497.
3. Gill, S.C. and von Hippel, P.H. (1989) *Anal. Biochem.* 182, 319–326.
4. Pace, C.N. et al. (1995) *Protein Science*, 4, 2411–2423.

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