

OncoStat Panel

Cat. No. NUC-063 **Lot. No.** (See product label)

SPECIFICATION

Product Overview	A panel of distinctly modified mononucleosomes assembled from recombinant human histones expressed in E. coli.
Source	E.coli
Description	A panel of distinctly modified 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 barcoded Widom 601 positioning sequence DNA. The OncoStat panel is comprised of a pool of 1 unmodified (H3.3) plus 7 histone H3.3 point mutations: H3.3K4M, H3.3K9M, H3.3K27M, H3.3G34R, H3.3G34V, H3.3G34W, H3.3K36M. Each distinctly modified nucleosome is distinguishable by a unique sequence of DNA ("barcode") at the 3' end that can be deciphered by qPCR or next-generation sequencing. Each of the 8 nucleosomes in the pool is wrapped by 2 DNA species, each containing a distinct barcode ("A" and "B", see SNAP-ChIP Manual) allowing for an internal technical replicate.
Form	Purified recombinant mononucleosomes, containing a mixture of 16 (1 unmodified plus 15 unique) H3 and H4 Purified recombinant mononucleosomes, containing a mixture of 8 (1 unmodified plus 7 point mutations) nucleosomes in 10mM sodium cacodylate, pH 7.5, 100 mM NaCl, 1 mM EDTA, 50% glycerol (w/v), 1x Protease Inhibitor cocktail, 100 ug/mL BSA, 10 mM β -mercaptoethanol. Molarity = 0.6 nM.
Molecular Mass	~199283.7 Da (average MW of all 8 nucleosomes)

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Applications

SNAP-ChIP OncoStats are highly purified recombinant mononucleosomes and are suitable for use as spike-in controls for ChIP reactions, for antibody specificity testing or for effector protein binding experiments. See manual for more information.

Storage

Stable for six months at -20°C from date of receipt.

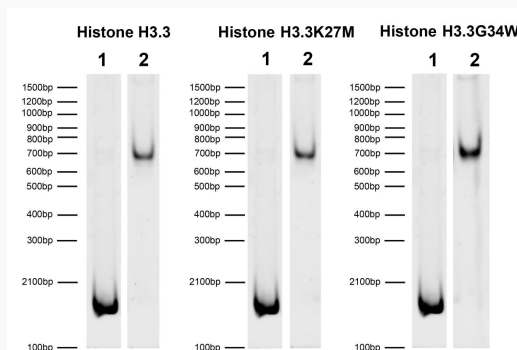
SNAP-ChIP OncoStat Panel

OncoStat Panel

H3.3	H3.3K4M	H3.3K9M	H3.3K27M
H3.3G34R	H3.3G34V	H3.3G34W	H3.3K36M

Schematic representation of nucleosomes comprising the OncoStat Panel: H3.3, H3.3K4M, H3.3K9M, H3.3K27M, H3.3G34R, H3.3G34V, H3.3G34W and H3.3K36M.

DNA Gel Data



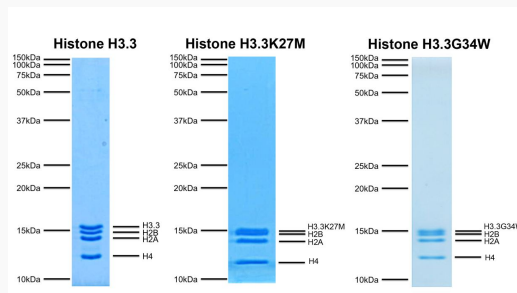
Representative images for SNAP-ChIP OncoStats resolved by native PAGE and stained with ethidium bromide to visualize DNA. Lane 1: Free 147bp DNA used in nucleosome assembly (100 ng). Lane 2: Intact nucleosomes (200 ng). Comparable experiments were performed for the entire OncoStat Panel.

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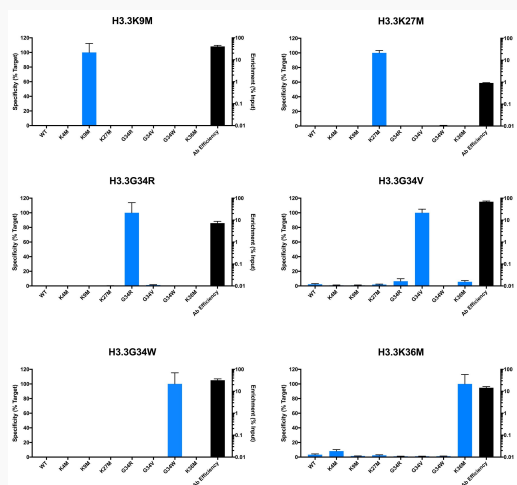
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Protein Gel Data



Representative Coomassie stained PAGE gel of SNAP-ChIP OncoStats (2 µg each) to demonstrate the purity of the histones in the preparation. Sizes of molecular weight markers and positions of the core histones (H2A, H2B, H3.3 and H4) are indicated. Comparable experiments were performed for the entire OncoStat Panel.

ChIP Data



Representative chromatin immunoprecipitation (ChIP) data using commercially available antibodies targeting single point mutations in histone H3.3. The antibodies were assayed in a native ChIP experiment with 3 µg antibody added to 3 µg HEK293 chromatin with the OncoStat Panel spiked-in prior to micrococcal nuclease digestion. Quantitative real-time PCR (qPCR) was used to measure recovery of

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duplicate DNA barcodes corresponding to each uniquely modified nucleosome in the panel (blue bars, X-axis). The black bars map to the log scale on the right y-axis and indicate the percentage of target immunoprecipitated relative to the input (a measure of the antibody efficiency). In each case, the SNAP-ChIP spike-in confirmed that the antibodies recovered the expected histone point mutation with high efficiency and specificity.

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