SNAP-ChIP® OncoStat Panel

Catalog No. 19-2001

Lot No. 21125002-02

Pack Size 20 µL

Product 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 mononucleosomes constitute a pool of 1 wild type (WT) unmodified 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 eight nucleosomes in the pool is wrapped by two DNA species, each containing a distinct barcode ("A" and "B", see SNAP-ChIP Manual) allowing for an internal technical replicate.

Formulation:

Purified recombinant mononucleosomes, containing a mixture of 8 (1 unmodified plus 7 point mutations) nucleosomes in 10 mM sodium cacodylate, pH 7.5, 100 mM NaCl, 1 mM EDTA, 50% glycerol (w/v), 1x Protease Inhibitor cocktail, 100 μ g/mL BSA, 10 mM β -mercaptoethanol. Average molarity = 0.6 nM. MW = ~199283.7 Da (average MW of all 8 nucleosomes).

Storage and Stability:

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

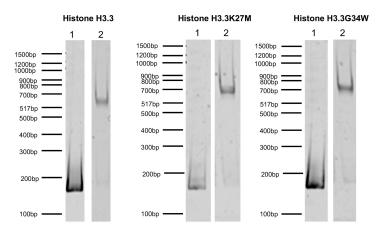
Application Notes:

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

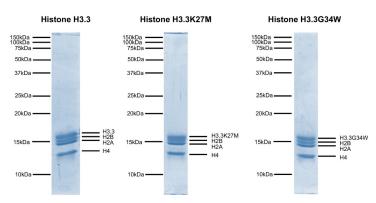
References Using this Product:

SNAP-ChIP is adapted from: Grzybowski AT et al (2015) Mol Cell 58: 886-889.



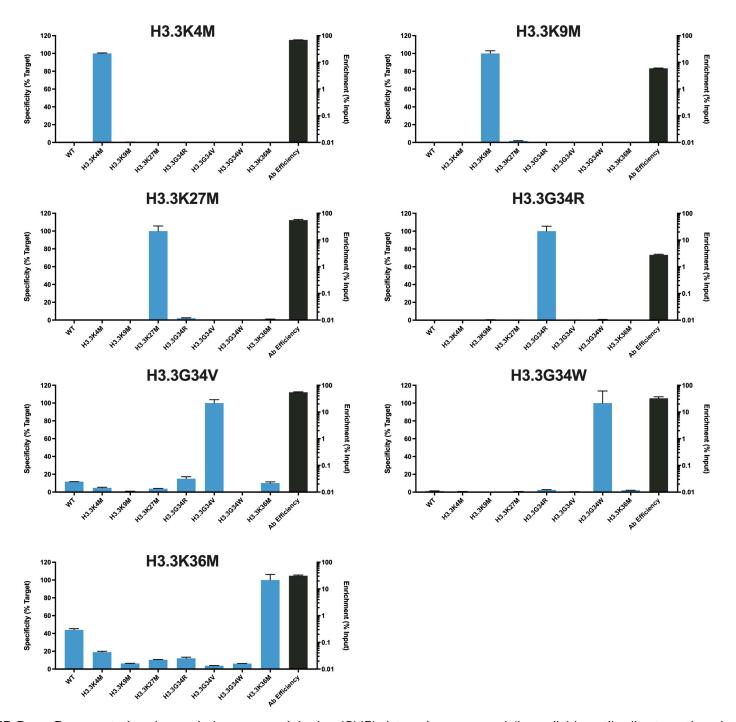


DNA Gel Data: Representative images for SNAP-ChIP OncoStat Panel nucleosomes resolved by native PAGE and stained with ethidium bromide to visualize DNA. Lane 1: Free 147 bp DNA used in nucleosome assembly (100 ng). Lane 2: Intact nucleosomes (200 ng). Comparable experiments were performed for the entire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



Protein Gel Data: Representative Coomassie stained PAGE gel of SNAP-ChIP OncoStat nucleosomes (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. For more information email techsupport@epicypher.com.

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



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 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. For more information, email techsupport@epicypher.com.