

# OncoStat Panel, SNAP-ChIP™ spike-in

Catalog Numbers A47343, A47355

Pub. No. MAN0019259 Rev. A.0

**WARNING!** Read the Safety Data Sheets (SDSs) and follow the handling instructions. Wear appropriate protective eyewear, clothing, and gloves. Safety Data Sheets (SDSs) are available from [thermofisher.com/support](http://thermofisher.com/support).

## Product Description

SNAP-ChIP™ spike-in, a proprietary method developed by EpiCypher™, is 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). Each histone is wrapped by 147 base pairs of barcoded Widom 601 positioning sequence DNA. The mononucleosomes constitute a pool of 1 unmodified histone plus 7 modified H3.3 histones (see Table 1). The modified H3.3 histones have point mutations. Each distinctly modified nucleosome is distinguishable by a unique sequence of DNA (the “barcode”) at the 3’ end that can be deciphered by qPCR or next-generation sequencing.

\* Histone H3.2 contains a Cys to Ala substitution at position 110.



## Contents and storage

Stable for one year at -20°C from date of receipt.

## Formulation

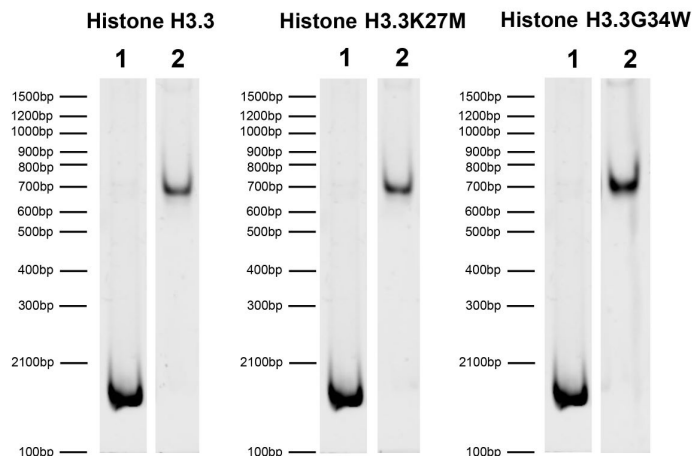
Purified recombinant mononucleosomes containing:

- A mixture of 8 histones in 10 mM sodium cacodylate, pH 7.5, 100 mM NaCl, 1 mM EDTA, 50% glycerol (w/v), 1x Protease Inhibitor cocktail, 100 µg/mL BSA, and 10 mM β-mercaptoethanol.
- Average molarity = 0.6 nM.
- MW = ~199283.7 Da (average MW of all 8 nucleosomes).

## Application notes

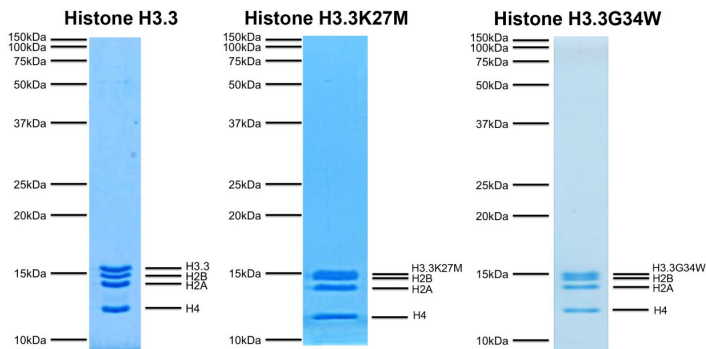
OncoStat Panel, SNAP-ChIP™ spike-in barcoded nucleosome standards are highly purified recombinant mononucleosomes and are suitable for use as spike-in controls for ChIP reactions, for antibody specificity testing, or for effector protein binding experiments. For more information, see *SNAP-ChIP Panels User Guide* (Pub. No. MAN0019284).

## Expected results



**Figure 1 DNA gel data**

Representative images for SNAP-ChIP™ OncoStats 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.



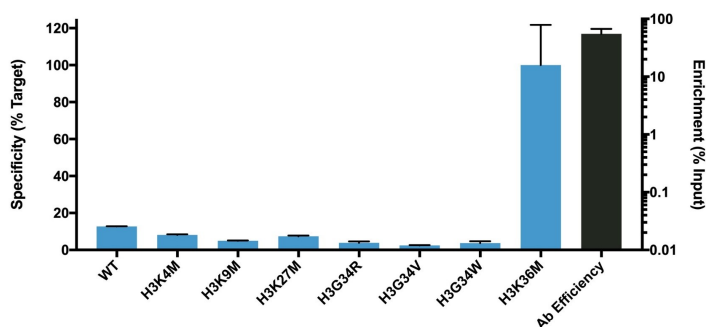
**Figure 2 Protein gel data**

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.

(Product # 711967) in ChIP. SNAP-ChIP panels consist of a pool of DNA-barcoded recombinant nucleosomes harboring unique oncogenic point mutations (on- and off-target) that are spiked-in to a ChIP reaction early in the workflow. The OncoStat panel includes H3.3 wild-type control as well as seven H3.3 mutants (K4M, K9M, K27M, G34R, G34V, G34W, K36M). Recovery of each unique DNA-barcoded nucleosome is quantified to determine how much of each species is immunoprecipitated in the ChIP reaction. H3K36M was tested in native ChIP with 3 µg K-652 cell chromatin and 3 µg antibody. Specificity (left Y-axis) was determined by quantitative real-time PCR (qPCR) to each modified nucleosome in the SNAP-ChIP™ OncoStat panel (X-axis). Black bar represents antibody efficiency (right Y-axis; log scale) and indicates percentage of the barcoded nucleosome target immunoprecipitated relative to Input. All bars represent mean ± SEM.

## References using this product

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



**Figure 3 Representative SNAP-ChIP data**

OncoStat Panel, SNAP-ChIP™ spike-in (Cat. No. A47343) was used to analyze the performance of recombinant polyclonal H3K36M antibody

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For descriptions of symbols on product labels or product documents, go to [thermofisher.com/symbols-definition](https://www.thermofisher.com/symbols-definition).

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