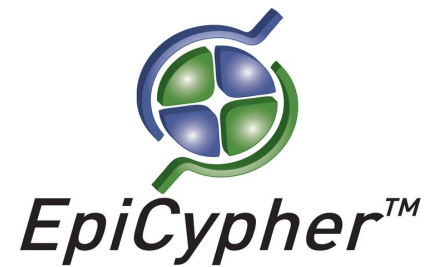


SNAP-ChIP K-MetStat Panel

Catalog No. 19-1001
Lot No. 17227001
Pack Size 20 μ l



Product Description:

A panel of distinctly modified mononucleosomes assembled from recombinant human histones expressed in *E. coli* (two each of histones H2A, H2B, H3 and H4; accession numbers: H2A-P04908; H2B-O60814; H3.1-P68431 or H3.2-Q71DI3*; H4-P62805) wrapped by 147 base pairs of barcoded Widom 601 positioning sequence DNA. The mononucleosomes constitute a pool of 1 unmodified plus 15 histone H3 or H4 post-translational modifications (PTMs, created by a proprietary semi-synthetic method): H3K4me1, H3K4me2, H3K4me3, H3K9me1, H3K9me2, H3K9me3, H3K27me1, H3K27me2, H3K27me3, H3K36me1, H3K36me2, H3K36me3, H4K20me1, H4K20me2 and H4K20me3. 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 16 nucleosomes in the pool is wrapped by 2 distinct DNA species, each containing a distinct barcode ("A" and "B", see SNAP-ChIP Manual) allowing for an internal technical replicate. * Histone H3.2 contains a Cys to Ala substitution at position 110.

Formulation:

Purified recombinant mononucleosomes, containing a mixture of 16 (1 unmodified plus 15 unique) H3 and H4 PTMs 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. Molarity = 0.6 nM. MW = ~199382.1 Da (average MW of all 16 nucleosomes).

Storage and Stability:

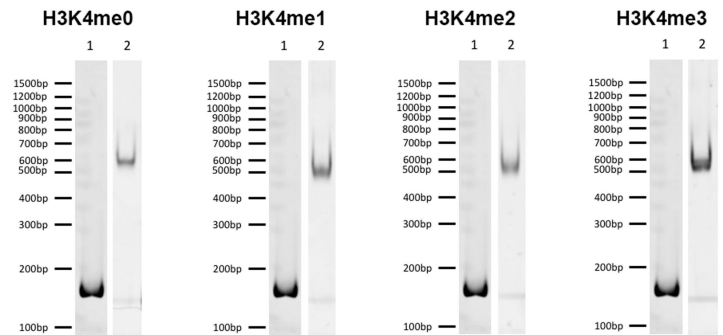
Stable for six months at -20°C from date of receipt.

Application Notes:

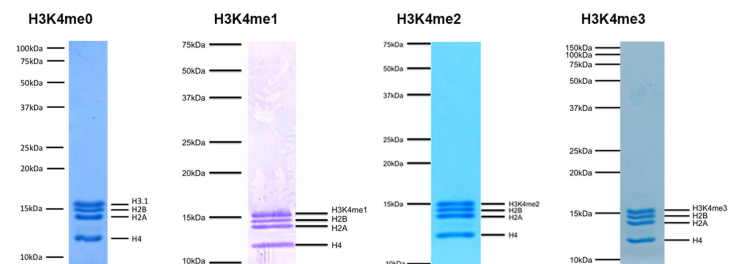
SNAP-ChIP K-MetStats are highly purified recombinant mononucleosomes and are suitable for use as spike-in controls for ChIP reactions, for antibody specificity testing or for effector protein binding experiments. See manual for more information.

References Using this Product:

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

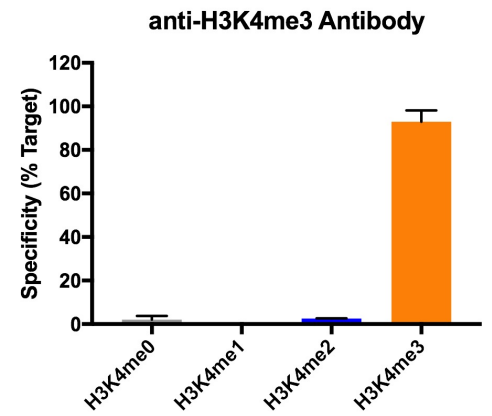
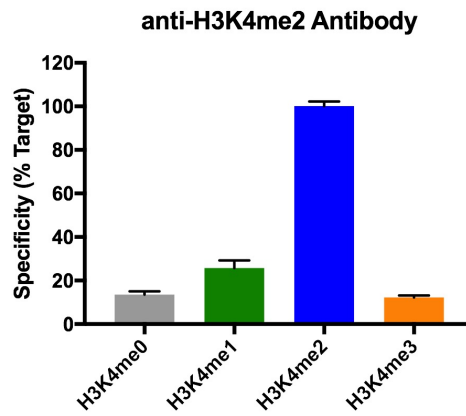
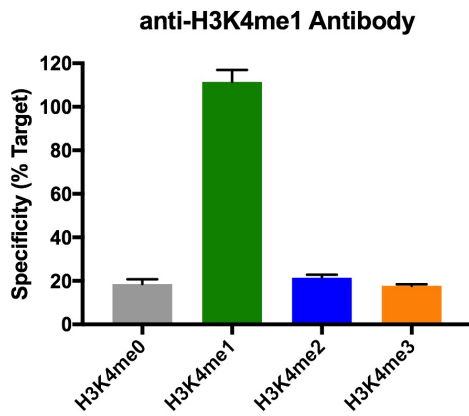


DNA Gel Data: Representative images for SNAP-ChIP K-MetStats (H3K4me0 = unmodified) resolved by native PAGE and stained with ethidium bromide to visualize DNA. **Lane 1:** Free 147bp DNA used in nucleosome assembly (100 ng). **Lane 2:** Intact nucleosomes (200 ng). Comparable experiments were performed for the entire K-MetStat Panel. Email techsupport@epicypher.com for more info.



Protein Gel Data: Representative Coomassie stained PAGE gel of SNAP-ChIP K-MetStats (2 μ g each; H3K4me0 = unmodified) to demonstrate the purity of the histones in the preparation. Sizes of molecular weight markers and positions of the core histones (H2A, H2B, H3 and H4) are indicated. Comparable experiments were performed for the entire K-MetStat 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 images for SNAP-ChIP K-MetStats (unmodified, H3K4me1, H3K4me2, H3K4me3) assayed in a chromatin immunoprecipitation (ChIP) experiment using commercially available ChIP-grade antibodies (3 μ g antibody added to 3 μ g HEK293 chromatin plus K-MetStats, n = 3). Quantitative real-time PCR (qPCR) to measure recovery of DNA barcodes corresponding to the nucleosomes indicated (X-axis; H3K4me0 = unmodified) pulled down with the specified antibody. Comparable experiments were performed for the entire K-MetStat Panel. For more information, email techsupport@epicypher.com.

N.B. -SNAP-ChIP is extremely useful for determining antibody cross-reactivity with off-target modifications. Any cross-reactivity observed above (e.g. anti-H3K4me1 & anti-H3K4me2) is not indicative of a failure in the SNAP-ChIP procedure, but represents off-target pull-down by the antibody. As seen above, only the H3K4me3 antibody exhibited the expected specificity.