Store at 4°C

Concanavalin A Magnetic Beads and Activation Buffer

#93569

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For Research Use Only. Not For Use In Diagnostic Procedures.

Products Included	Product #	Quantity	Storage Temp
Concanavalin A Magnetic Beads	82307	240 µl	4°C
Concanavalin A Bead Activation Buffer	91275	5 ml	4°C

See www.cellsignal.com for individual component applications, species cross-reactivity, dilutions and additional application protocols.

Description: The Concanavalin A Magnetic Beads and Activation Buffer kit provides enough reagents to support 24 CUT&RUN assays. This product is tested and validated using the CUT&RUN Assay Kit #86652. This product should be stored at 4°C. Please do not freeze the Concanavalin A Magnetic Beads!

Background: Like the chromatin immunoprecipitation (ChIP) assay, Cleavage Under Targets and Release Using Nuclease (CUT&RUN) is a powerful and versatile technique used for probing protein-DNA interactions within the natural chromatin context of the cell (1-4). CUT&RUN provides a rapid, robust, and true low cell number assay for detection of protein-DNA interactions in the cell. Unlike the ChIP assay, CUT&RUN is free from formaldehyde cross-linking, chromatin fragmentation, and immunoprecipitation, making it a much faster and more efficient method for enriching protein-DNA interactions and identifying target genes. CUT&RUN can be performed in less than one day, from live cells to purified DNA, and has been shown to work with as few as 500-1000 cells per assay (1,2). Instead of fragmenting all of the cellular chromatin as done in ChIP, CUT&RUN utilizes an antibody-targeted digestion of chromatin, resulting in much lower background signal than seen in the ChIP assay. As a result, CUT&RUN requires only 1/10th the sequencing depth that is required for ChIP-seq assays (1,2). Finally, the inclusion of simple spike-in control DNA allows for accurate quantification and normalization of target-protein binding that is not possible with the ChIP method. This provides for effective normalization of signal between samples and between experiments.

Storage: Store at 4°C. Do not freeze the Concanavalin A Magnetic Beads! *This product is stable for at least 12 months.*

Directions for Use: For the CUT&RUN assay, we recommend using 10 µl Concanavalin A Magnetic Beads per reaction. Before use, the Concanavalin A Magnetic Beads should be washed 2 times with 10X volume of Concanavalin A Bead Activation Buffer and resuspended in a volume of Concanavalin A Bead Activation Buffer equal to the initial volume of bead suspension. Activated beads should be used within one day.

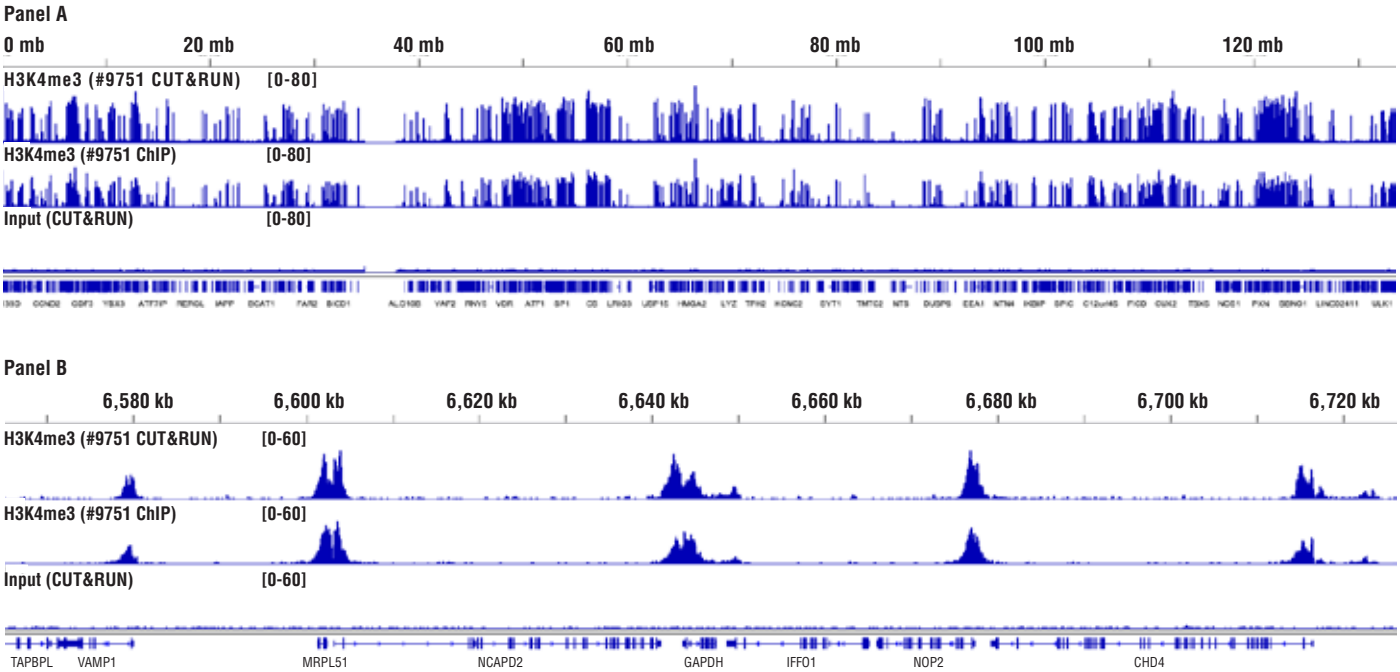
Background References:

(1) Skene, P.J. and Henikoff, S. (2017) *Elife* 6, pii: e21856. doi: 10.7554/eLife.21856.

(2) Skene, P.J. et al. (2018) *Nat Protoc* 13, 1006-19.

(3) Meers, M.P. et al. (2019) *Elife* 8, pii: e46314. doi: 10.7554/eLife.46314.

(4) Meers, M.P. et al. (2019) *Mol Cell* 75, 562-575.e5.



CUT&RUN and ChIP assays were performed with HCT 116 cells and Tri-Methyl-Histone H3 (Lys4) (C42D8) Rabbit mAb #9751. DNA Libraries were prepared using SimpleChIP® ChIP-seq DNA Library Prep Kit for Illumina® #56795. Panel A compares enrichment of H3K4me3 across chromosome 12 (upper), while Panel B compares enrichment at the GAPDH gene (lower), a known target of H3K4me3. The input tracks are from the CUT&RUN input sample.

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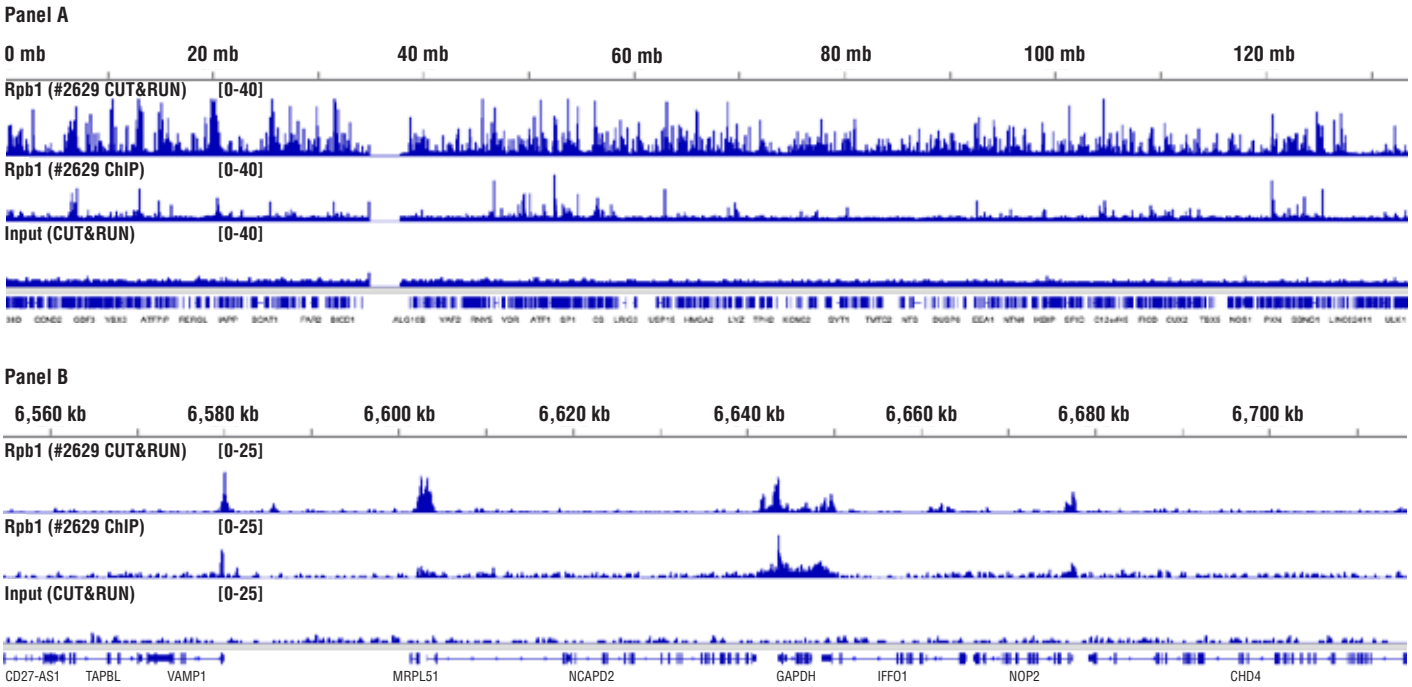
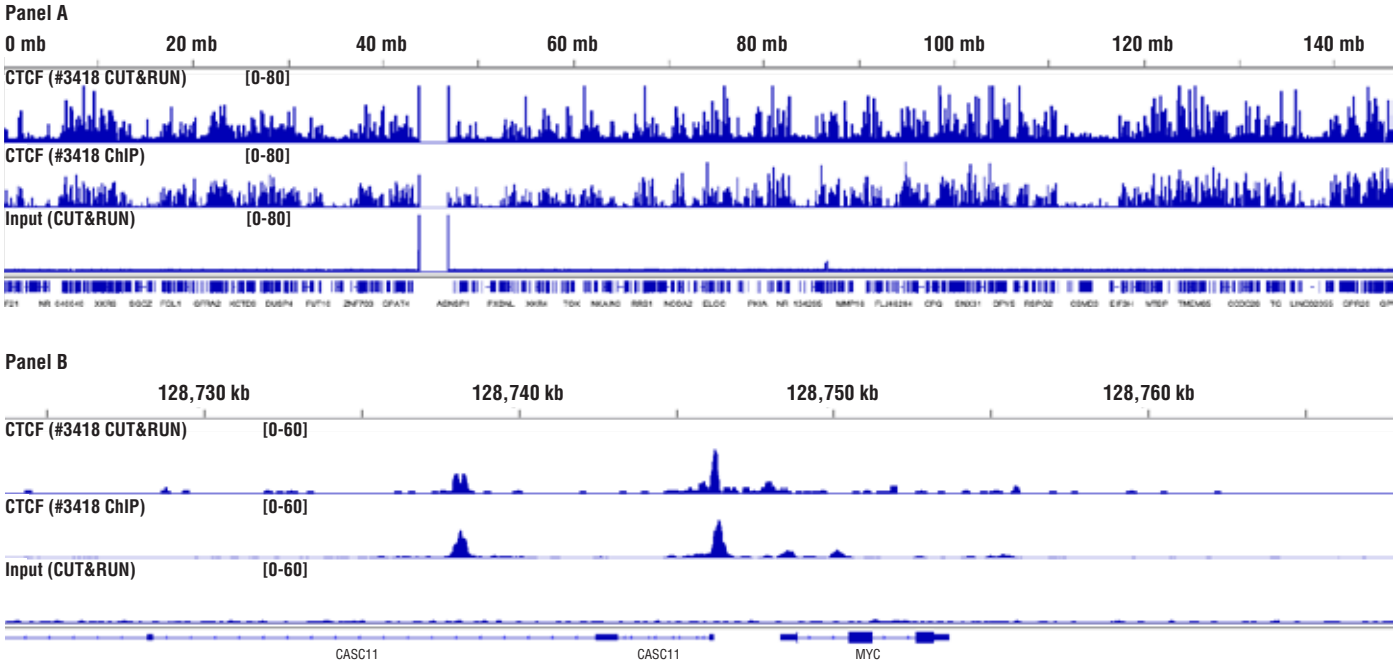
Applications: W—Western **IP**—Immunoprecipitation **IHC**—Immunohistochemistry **ChIP**—Chromatin Immunoprecipitation **IF**—Immunofluorescence **F**—Flow cytometry **E-P**—ELISA-Peptide **Species Cross-Reactivity: H**—human **M**—mouse **R**—rat **Hm**—hamster **Mk**—monkey **Mi**—mink **C**—chicken **Dm**—D. melanogaster **X**—Xenopus **Z**—zebrafish **B**—bovine **Dg**—dog **Pg**—pig **Sc**—S. cerevisiae **Ce**—C. elegans **Hr**—Horse **All**—all species expected Species enclosed in parentheses are predicted to react based on 100% homology.



CUT&RUN and ChIP assays were performed with HeLa cells and Rpb1 CTD (4H8) Mouse mAb #2629. DNA Libraries were prepared using SimpleChIP® ChIP-seq DNA Library Prep Kit for Illumina® #56795. Panel A compares enrichment of Rpb1 across chromosome 12 (upper), while Panel B compares enrichment at the GAPDH gene (lower), a known target of Rpb1. The input tracks are from the CUT&RUN input sample.



CUT&RUN and ChIP assays were performed with HCT 116 cells and CTCF (D31H2) XP® Rabbit mAb #3418. DNA Libraries were prepared using SimpleChIP® ChIP-seq DNA Library Prep Kit for Illumina® #56795. Panel A compares enrichment of CTCF across chromosome 8 (upper), while Panel B compares enrichment at the MYC gene (lower), a known target of CTCF. The input tracks are from the CUT&RUN input sample.