

Store at
-20°C

100X Spermidine

#27287

1.3 mL

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

Description: 100X Spermidine is offered to conveniently provide additional spermidine for use with the CUT&RUN Assay Kit #86652 or the CUT&Tag Assay Kit #77552. Both CUT&RUN Assay Kit #86652 and CUT&Tag Assay Kit #77552 provide all the reagents required for performing the recommended number of Cleavage Under Targets & Release Using Nuclease (CUT&RUN) and Cleavage Under Targets & Tagmentation (CUT&Tag) assays. However, there are instances where extra spermidine is desired. This product can be added to the Antibody Binding Buffer (CUT&RUN, CUT&Tag) #15338 or 10X Wash Buffer (CUT&RUN, CUT&Tag) #31415 right before use, or be used in other assays as needed. Please keep on ice during use and store at -20°C when finished for the day.

Background: Like the chromatin immunoprecipitation (ChIP) assay, Cleavage Under Targets and Release Using Nuclease (CUT&RUN) and Cleavage Under Targets and Tagmentation (CUT&Tag) are powerful and versatile techniques used for probing protein-DNA interactions within the natural chromatin context of the cell (1-7). 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 500-1,000 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 signals between samples and between experiments. CUT&Tag has many of the same advantages as the CUT&RUN assay in that it provides a rapid, robust, and true low cell number protocol for detection of protein-DNA interactions in the cell. In addition, the CUT&Tag assay adds an in situ adaptor DNA ligation step carried out by the pAG-Tn5 enzyme, in which an adaptor DNA is ligated directly to antibody-targeted chromatin DNA fragments in the cell. As a result, subsequent DNA library preparation is much faster and easier than library preparation following the CUT&RUN assay, free from DNA end repair, A-tailing, and adaptor ligation in vitro. CUT&Tag works very well for analyzing histone modifications, in addition to mapping some transcription factors and cofactors binding.

Storage: Store 100X Spermidine at -20°C. This product is stable for at least 12 months.

Please visit www.cellsignal.com for a complete listing of recommended companion products.

Directions for Use: For the CUT&RUN or CUT&Tag assay, use this product as a 100X stock, as directed in the CUT&RUN Assay Kit #86652 or CUT&Tag Assay Kit #77552 protocols.

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.

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