CUTANA™ CUT&RUN and CUT&Tag epigenomic mapping assays for agricultural research

Kelsey E. Noll[1], Katherine Novitzky[1], Matthew R. Marunde[1], Danielle N. Maryanski[1], Catherine Smith[1], Allison Hickman[1], Kateřina Adamusová[2], Karina Chalenko[2], Swetha Chenna[2], Quentin Thomas[2], Jinghan Liu[2], Andrea L. Johnstone[1], Keith E. Maier[1], Ellen Weinzapfel[1], Irina K. Popova[1], Bryan J. Venters[1], Keli L. Rodriguez[1], Carolina P. Lin[1], Ashton Gladney[1], Rachel Watson[1], Zachary B. Gillespie[1], Augustus Adeleke[1], Saarang Gopinath[1], Leslie Lewis[1], Hannah Willis[1], Lu Sun[1], Marcus A. Cheek[1], Matthew J. Meiners[1], James R. Bone[1], Martis W. Cowles[1], Zu-Wen Sun[1], Sebastian Marquardt[2], Michael-Christopher Keogh[1]

[1] EpiCypher Inc., Durham, NC, USA; [2] Department of Plant and Environmental Sciences, Copenhagen Plant Science Centre, University of Copenhagen, Denmark

Epigenomic mapping provides key insights into gene regulation, but many commonly used approaches are not sufficient

- Many genomic strategies for cellular profiling focus on transcription; however, RNA-seq reveals the **outcomes** – not driving **mechanisms**
- **Epigenomics is the solution**: mapping the location of chromatin features provides mechanistic insights that are central to cell fate and function
- Existing epigenomic technologies (e.g., ChIP-seq) lack standardized controls, require high cell inputs, are low-throughput, and have high costs

CUTANA™ CUT&RUN and CUT&Tag chromatin mapping assays are highly enabling to advance epigenetic studies

Compared to ChIP-seq, CUT&RUN and CUT&Tag generate data with >100-fold fewer cells and >10-fold reduced sequencing depth, with >3-fold cost improvement per reaction

- CUT&RUN and CUT&Tag are next-generation immunotethering-based mapping approaches
- Nuclei extraction and genomic mapping can be performed across separate days and worksites

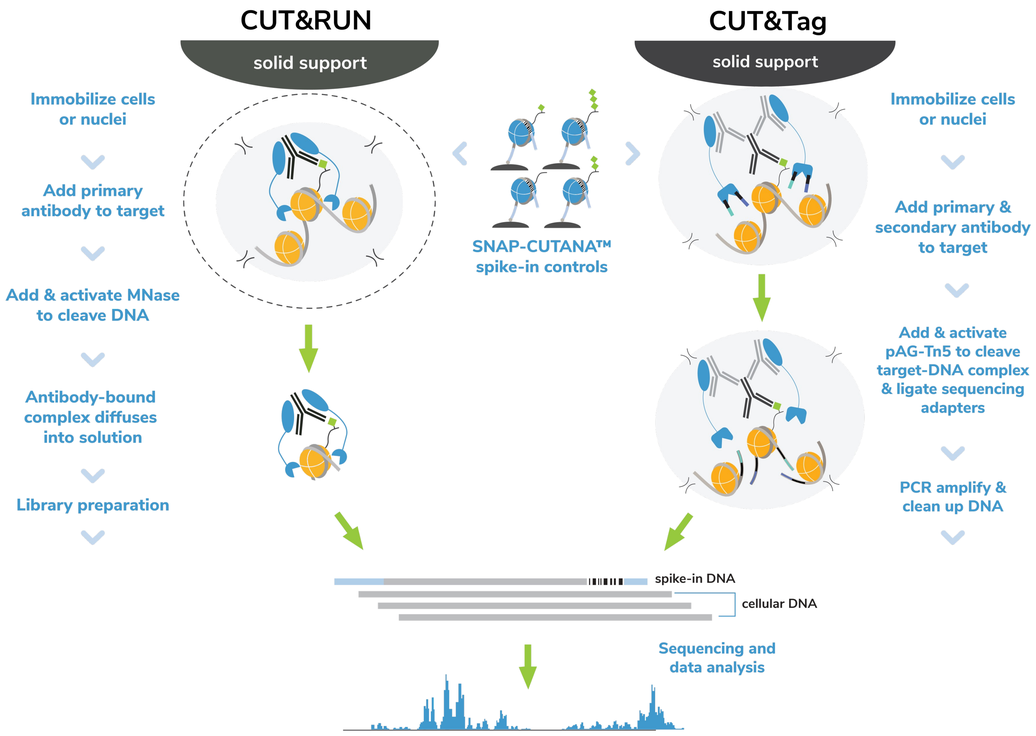

Figure 1: CUTANA™ CUT&RUN and CUT&Tag workflows for chromatin mapping

Defined nucleosome spike-in controls are necessary for reliable epigenomic mapping and data analysis

SNAP-CUTANA™ nucleosome spike-in controls are specifically designed for compatibility with CUTANA™ CUT&RUN and CUT&Tag assays, and are essential reagents to enable:

1. Antibody validation
2. Technical monitoring
3. Quantitative normalization

>70% **of histone PTM antibodies are unfit for genomic mapping** due to poor specificity and/or low efficiency
search your antibody of interest at www.chromatinantibodies.com

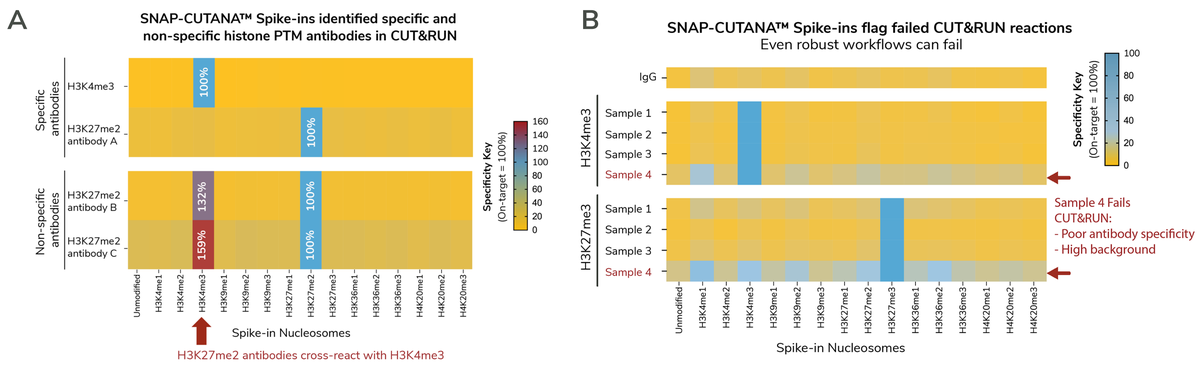

Figure 2: SNAP-CUTANA™ spike-in control heatmaps highlighting applications to **(A)** validate antibody specificity in-assay, and **(B)** monitor technical performance for sample quality control.

Automated CUTANA™ assays enable genomic mapping at scale

Platform comparison	ChIP-seq	CUTANA™ CUT&RUN and CUT&Tag	Implications for automation
Sample input	Fragmented chromatin	Intact cells or nuclei	Streamlined workflow
Required cells	>1 million	500k - 5k	More data from precious samples
Defined controls	Uncommon	SNAP-CUTANA spike-ins	Standardized protocols
Seq depth (reads)	>30 million (mammalian)	3-5 million (mammalian)	Greater multiplexing
Assay cost (per reaction)	~$225	~$72	70% cost savings
Background	High	Low	Better data quality
Experimental throughput	Low	High	Compatible with 96-well plates

Automation halves hands-on time and increases throughput >8-fold

- End-to-end optimization: sample prep to library QC
- Reduced variance and reaction volumes with 96-well liquid handling
- Standardized workflow for native and cross-linked cells and nuclei
- Buffer optimization for improved sample handling
- AutoCUT&RUN assays generate reliable signal down to 5,000 mammalian cells
- AutoCUT&Tag assays in active development

CUTANA™ CUT&RUN and CUT&Tag assays are enabling for next-generation agricultural research

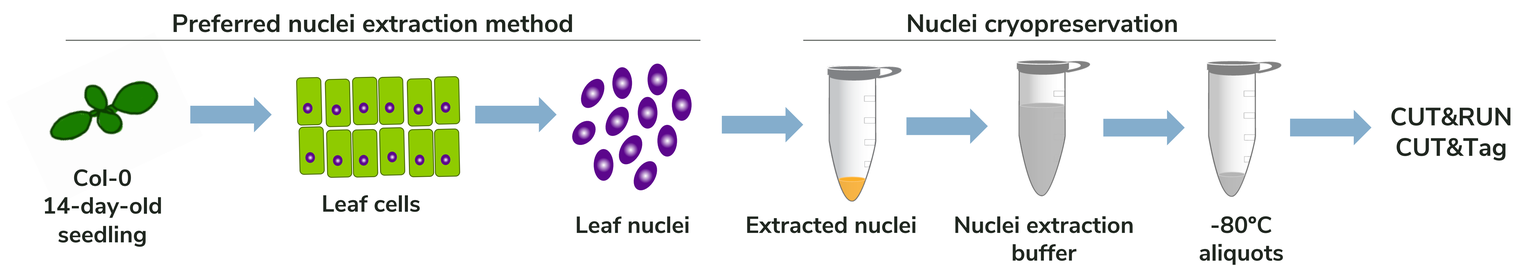

Figure 4: Workflow for sample processing of *Arabidopsis thaliana* nuclei upstream of CUTANA™ CUT&RUN and CUT&Tag assays. Cryopreservation in EpiCypher nuclei extraction buffer (Cat. No. 21-1026) is critical to preserve plant nuclei integrity and enables flexibility to perform the assay across multiple days and/or collaborative sites.

- **Both CUT&RUN and CUT&Tag are compatible with plant cells**
- Preliminary experiments indicate CUT&Tag may generate higher signal-to-noise

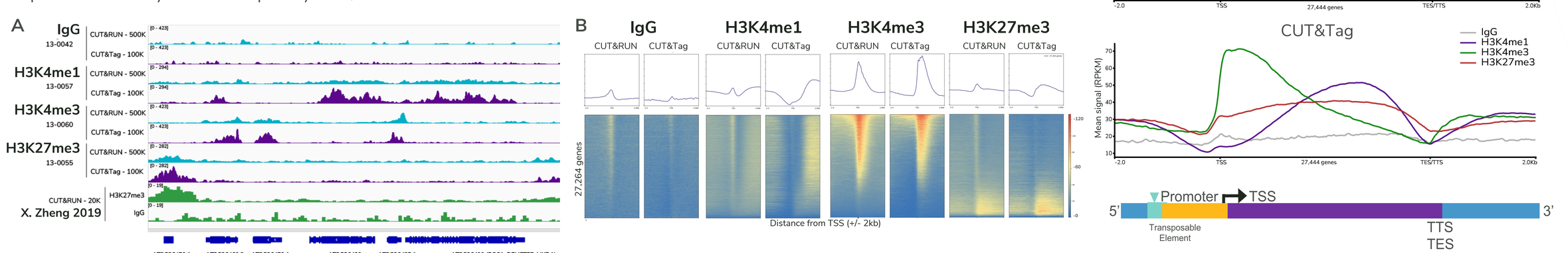

Figure 5: CUTANA™ CUT&RUN and CUT&Tag assays map epigenomic landscape of *Arabidopsis thaliana* seedlings. (**A**) IGV tracks of histone PTMs; H3K27me3 aligns with previously published tracks (Zheng, 2019). (**B**) Heatmaps show PTM signal relative to transcription start sites (TSS), genes ordered by H3K4me3 signal. (**C**) Metagene plots show mean signal for each PTM relative to annotated genomic features.

Download poster