Advances in chromatin remodeling research

EpiCypher.

Chromatin remodeling complexes are crucial regulators of chromatin structure and gene expression. An estimated 20% of human cancers contain mutations in the SWI/SNF remodeling complex, making these proteins important drug targets. EpiCypher is at the forefront of epigenetics technology development, providing new tools to transform chromatin remodeling research.

FAMILY	COMPLEX	ENZYME	DISEASES
SWI/SNF	BAF	SMARCA2/BRM or SMARCA4/BRG1	Neurodevelopmental disorders, including autism spectrum disorder Coffin-Siris syndrome Cancer: rhabdoid tumors, synovial sarcoma, non-small cell lung carcinoma
	PBAF	SMARCA4/BRG1	
ISWI	NURF	SMARCA1/SNF2L	Intellectual disability, developmental delay Cancer: melanoma, lung, neuroblastoma
	ACF	SMARCA5/SNF2H	
	CHRAC		
CHD	NuRD	CHD3, CHD4	Neurodevelopmental disorders, including autism spectrum disorder CHARGE syndrome Cancer: neuroblastoma, prostate Dermatomyositis
	CHD1	CHD1	
INO80	INO80	INO80	Neurodevelopmental disorders, microcephaly, Alzheimer's disease Cancer: melanoma, thyroid, cervical Congenital heart disease
	TIP60	p400	
	SCRAP	SRCAP	

TABLE 1

Summary of diseases linked to chromatin remodeling complexes. List is not exhaustive. For more information, see Goodwin et al. (2018), Centore et al. (2020), Li et al. (2021), Alendar et al. (2021), and Poli et al. (2017).

EpiDyne™ chromatin remodeling assays

Chromatin remodelers have been challenging to target for therapeutic development, as there were no assays to directly monitor remodeling activity. The EpiDyne platform was uniquely created to address this problem, leveraging recent advances in biochemistry to advance the study of remodeling complexes.

- Many readouts available: restriction enzyme accessibility, FRET, and more
- Quantitative analysis on a nucleosome substrate
- Only commerical provider of enzymatically active SMARCA4/BRG1 and SMARCA2/BRM
- HTS-compatible for drug discovery

Enzyme titration

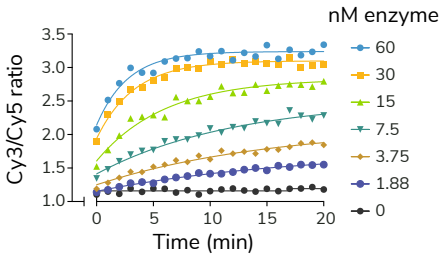

FIGURE 1

Ultra-sensitive readout of SWI/SNF remodeling activity using EpiDyne substrates and enzymes. SMARCA4 enzyme was titrated against EpiDyne-FRET nucleosomes. Remodeling activity was determined by the ratio of Cy3/Cy5 at varying time points.

Z' determination

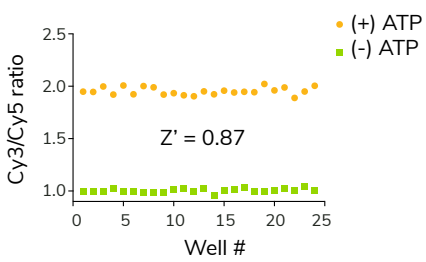

FIGURE 2

EpiDyne is suitable for high-throughput screening. SMARCA4 was incubated in triplicate with EpiDyne-FRET nucleosomes, with (+) and without (-) ATP. Z' was calculated as an indicator of consistency and reliability for HTS. Reactions contained DMSO to mimic HTS.

Inhibitor dose response

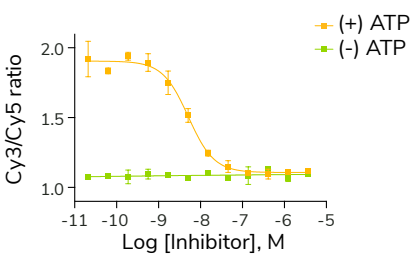

FIGURE 3

Characterization of a SWI/SNF inhibitor using EpiDyne assays. The Novartis inhibitor BRM014/Compound 14 (Papillon et al. 2018; Jagani et al. 2019) displayed dose-dependent inhibition of chromatin remodeling activity as indicated by a reduction in Cy3/Cy5 FRET.

CUT&RUN assays for chromatin remodelers

The genome-wide localization of chromatin remodeler proteins is essential for understanding their function in disease but has been obscured by the stringent salt wash steps associated with ChIP-seq. CUTANA™ CUT&RUN provides a robust, low-cost approach to functionally characterize chromatin remodeling complexes in vivo. We have used our CUTANA™ CUT&RUN kits and antibodies to map major classes of chromatin remodeling enzymes, demonstrating high quality resolution (Figure 4).

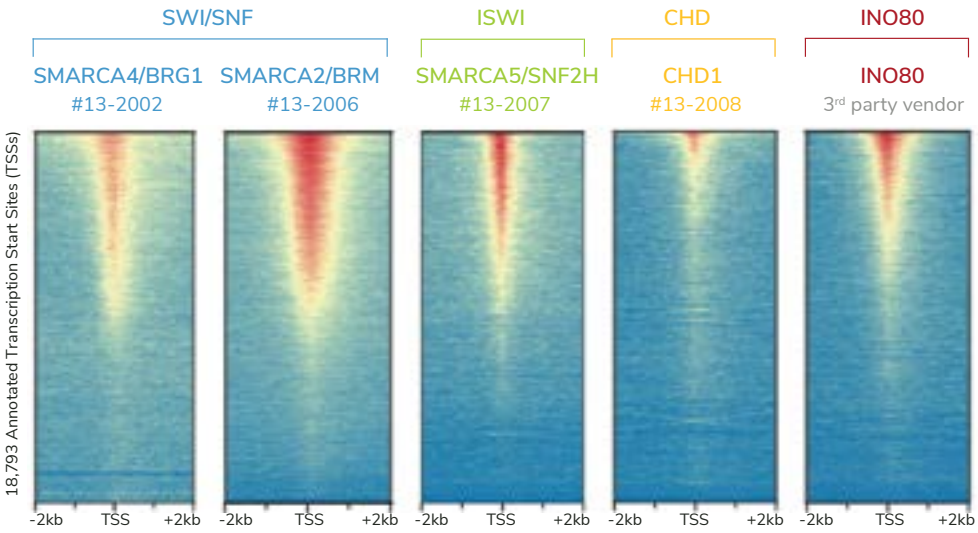

FIGURE 4

CUT&RUN generates reliable profiles with high signal-to-noise for diverse chromatin remodeling enzymes. Reactions were performed using 500,000 K562 cells and the CUTANA CUT&RUN Kit. Heatmaps are aligned to transcription start sites (TSS) and ranked by peak signal intensity.

CUTANA™ CUT&RUN antibodies

EpiCypher offers extensively validated CUTANA CUT&RUN antibodies to key chromatin remodeling enzymes, including the high-value drug target SMARCA4/BRG1. Each antibody is rigorously lot-tested in CUT&RUN, and genome-wide distribution is compared with known overlapping signaling pathways for unprecedented biological validation. Our final antibodies generate reliable profiles with high signal-to-noise, providing a powerful approach to study chromatin remodelers in vivo.

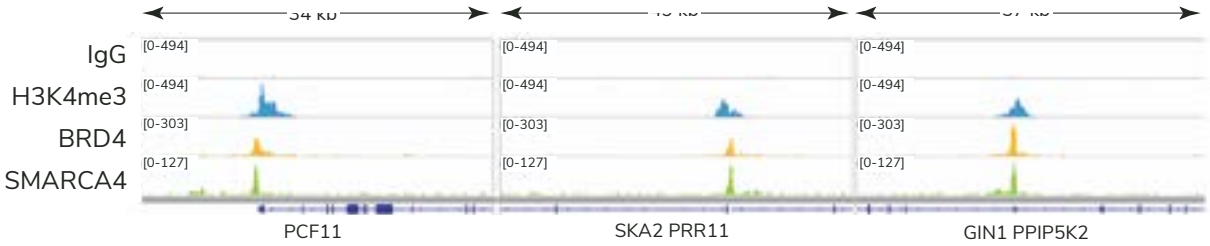

FIGURE 5 **SMARCA4/BRG1 was mapped in K562 cells using our CUTANA™ CUT&RUN antibody and CUT&RUN kit.** To validate this antibody, we compared SMARCA4/BRG1 maps with CUT&RUN profiles for related targets, including H3K4me3 (denotes TSS) and BRD4 (interacts with SMARCA4/BRG1). IgG included as negative control.