

Resource Summary Report

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CUT&RUN: Targeted in situ genome-wide profiling with high efficiency for low cell numbers

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Protocol Information

URL: <https://dx.doi.org/10.17504/protocols.io.mgjc3un>

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Group: Human Cell Atlas Method Development Community

Summary: Cleavage Under Targets and Release Using Nuclease (CUT&RUN) is an epigenomic profiling strategy in which antibody-targeted controlled cleavage by micrococcal nuclease releases specific protein-DNA complexes into the supernatant for paired-end DNA sequencing. As only the targeted fragments enter into solution, and the vast majority of DNA is left behind, CUT&RUN has exceptionally low background levels. CUT&RUN outperforms the most widely used Chromatin Immunoprecipitation (ChIP) protocols in resolution, signal-to-noise, and depth of sequencing required. In contrast to ChIP, CUT&RUN is free of solubility and DNA accessibility artifacts and can be used to profile insoluble chromatin and to detect long-range 3D contacts without cross-linking. Here we present an improved CUT&RUN protocol that does not require isolation of nuclei and provides high-quality data starting with only 100 cells for a histone modification and 1000 cells for a transcription factor. From cells to purified DNA CUT&RUN requires less than a day at the lab bench. In summary, CUT&RUN has several advantages over ChIP-seq: (1) The method is performed in situ in non-crosslinked cells and does not require chromatin fragmentation or solubilization; (2) The intrinsically low background allows low sequence depth and identification of low signal genomic features invisible to ChIP; (3) The simple procedure can be completed within a day and is suitable for robotic automation; (4) The method can be used with low cell numbers compared to existing methodologies; (5) A simple spike-in strategy can be used for accurate quantitation of protein-DNA interactions. As such, CUT&RUN represents an attractive replacement for ChIPseq, which is one of the most popular methods in biological research.

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