Abstract for AACR 2019

TAPS: A bisulfite-free, base-resolution and quantitative sequencing method for cytosine modifications

Yibin Liu, Paulina Siejka, Gergana Velikova, Ying Bi, Marketa Tomkova, Benjamin Schuster-Boeckler and Chunxiao Song

Aberrant DNA methylation changes are powerful prognostic and predictive biomarkers in cancer diagnosis and treatment. For decades bisulfite sequencing has been the gold standard for DNA methylation analysis. However, this method relies on harsh deamination reaction of unmodified cytosine to uracil which degrades the majority of DNA and generates sequencing libraries with low complexity. Those drawbacks limit the applications on low-input samples and lead to poor sequencing quality, low mapping rates, uneven genome coverage and increased sequencing cost. Here, we present a novel bisulfite-free and base-resolution sequencing method, TET Assisted Pyridine borane Sequencing (TAPS), for direct detection of 5-methylcytosine (5mC) and 5-hydroxymethylcytosine (5hmC) without affecting unmodified cytosine. We utilised TET enzymes to readily oxidize 5mC and 5hmC to 5-carboxylcytosine (5caC), which was then converted to dihydrouracil by pyridine borane via a previously unknown reductive decarboxylation/deamination reaction. This non-destructive method enables high-throughput sequencing with nanoscale input materials (genomic DNA and cell-free DNA) and preserve DNA up to 10 kbs long which could allow long read sequencing. We demonstrated that TAPS could directly replace bisulfite sequencing with higher mapping rates, more even coverage and lower sequencing costs, and enable more comprehensive, cheaper and faster methylome analyses. This would be a revolutionary tool for epigenetic study and facilitate both academic research and clinical diagnostics.