

Direct Methylation Sequencing (dMS)

Renew Biotechnologies Service

Summary

Direct Methylation Sequencing (dMS) is Renew's cell-free DNA (cfDNA) sequencing service optimized for native methylation analysis. Combining proprietary library preparation with native Oxford Nanopore sequencing, dMS enables genome-wide methylation profiling from circulating cfDNA without bisulfite conversion or PCR amplification while simultaneously capturing native sequence information from the same dataset.

Designed for liquid biopsy applications, dMS supports comprehensive cfDNA methylation analysis for biomarker discovery, tissue-of-origin research, disease monitoring, and translational development.

Key Features & Benefits

- **Native cfDNA Methylation Profiling:** Capture native 5mC and 5hmC methylation without bisulfite conversion or PCR amplification.
- **Integrated Multiomic Analysis:** Generate sequence and methylation information from the same cfDNA dataset.
- **Liquid Biopsy Optimized:** Proprietary workflow designed specifically for fragmented circulating cell-free DNA.
- **Comprehensive Methylation Analysis:** Profile methylation patterns across the genome beyond predefined CpG targets.

Ideal Applications

- Liquid biopsy biomarker research
- Tissue-of-origin analysis
- Disease monitoring and longitudinal studies
- Translational cfDNA methylation research

Protocol Overview

Library Preparation: Proprietary Renew Direct Methylation Sequencing workflow

Sequencing Platform: Oxford Nanopore PromethION

Workflow

- 1 cfDNA Extraction:** Cell-free DNA is isolated from plasma or other validated biofluids.
- 2 Library Preparation:** Samples undergo Renew's proprietary native cfDNA library preparation and adapter ligation workflow.
- 3 Native Long-Read Sequencing:** Libraries are sequenced on the Oxford Nanopore PromethION platform.
- 4 Bioinformatic Processing:** Sequencing data are processed through automated cfDNA methylation analysis workflows.
- 5 Data Delivery:** Results are delivered through the customer portal.

Sample Requirements	
Parameter	Requirement
Sample Type	Cell-Free DNA (cfDNA)
Minimum Input	5 ng
Recommended Input	50 ng

Sequencing Performance	
Coverage and sequencing yield depend on cfDNA quality and project objectives.	
Metric	Typical Performance
Samples per Flow Cell	24
Genome Coverage	Project dependent
Epigenetic Signals	Native 5mC and 5hmC

Throughput and Capacity

Sequencing depth is tailored to study objectives, multiplexing, sample quality, and desired analytical sensitivity.

Data Output

File Format

Renew provides standard sequencing files, quality reports, and analysis outputs for downstream review and interpretation.

Bioinformatics Outputs

Sequencing data are processed through standardized and proprietary methylation analysis workflows to generate native CpG methylation measurements and quality metrics.

Standard Methylation Analysis

modkit extract	read-level CpG methylation calls
modkit pileup	aggregated methylation levels per CpG site

File	Description
POD5	Raw nanopore sequencing data, available upon request for an additional fee.
BAM	Aligned sequencing reads used for downstream telomere, methylation, and variant-aware analysis.

File	Description
CH3	Renew's base modification output format, designed to organize methylation and other epigenetic calls for downstream use.
QC Report	Summary of sequencing metrics, run statistics, sample-level performance, and quality indicators.

Renew's Methylation Data Ecosystem

Renew's proprietary CH3 file format and ModSeqR analysis package simplify the management and downstream analysis of native long-read methylation data.

CH3: Efficient Native Methylation Storage

CH3 is Renew's compressed file format for base-resolution methylation data, reducing file size by up to 95% while preserving information needed for efficient downstream storage, transfer, and analysis of methylomes.

ModSeqR: Native Methylation Analysis in R

ModSeqR is Renew's open-source R package for analyzing CH3 data, providing quality control, regional and differential methylation analysis, plus array-compatible conversion for minfi, DMRcate, and chAMP workflows using native methylation.

Data Delivery

Delivery Option	Description
Client Web Portal	Secure browser-based access for manual download on Mac, Linux, or Windows.
AWS S3 Transfer	Automated AWS-to-AWS transfer option for teams managing large datasets in cloud environments.
Command Line Script	Manual command-line download option for Mac and Linux users who prefer scripted file retrieval.

Turnaround Time

Results appear in Renew's client portal within 14-21 days from sample receipt**.

**May vary with project scope, sample quality, and sequencing depth and bioinformatic requirements.

Service Inclusions

Each project includes dedicated scientific and project management support spanning study design through data delivery.

- Experimental design consultation
- Proprietary cfDNA library preparation
- Standardized sequencing QC
- Live project tracking
- Standard methylation analysis
- Post-project technical consultation
- Flexible data delivery

Contact and Pricing

Contact us for more information or request a quote

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