REDUCED REPRESENTATION BISULFITE SEQUENCING (RRBS)

Empowering Australian Genomics

Methylation beyond the human genome

Reduced Representation Bisulfite Sequencing (RRBS) is a comprehensive analysis method and an economical alternative to whole genome bisulfite sequencing - requiring up to approximately 50 fold less sequencing.
It provides strand specific single-base resolution of DNA methylation (5 methylC) across a broader genomic span than predesigned arrays.

HOW IT WORKS

Genomic DNA is digested with the methylation insensitive enzyme MspI with the recognition site CCGG. Sequencing adapters with cohesive-ends (or sticky-ends) are ligated directly to the digested DNA. Following adapter ligation the DNA is bisulfite treated converting un-methylated cytosines to uracil and then amplified. The resulting fragments are enriched for potential CpG methylation sites.

Similar to exome-sequencing for mutation discovery, the RRBS protocol enriches for some of the most interesting target regions and thereby achieves a reduction in sequencing cost compared to whole genome bisulfite sequencing.

The method is flexible in design - it is suitable for both model and non-model species with a reference genome, and delivers a high-throughput method that is well suited to genome-wide DNA methylation studies.

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SUPERIOR CAPABILITIES FOR DNA METHYLATION STUDIES

- Superior coverage– potentially millions of CpGs
- High throughput - 96 samples processed in one experiment
- Cost-efficient – multiplexing samples economizes sequencing
- High efficiency and minimal bias - method uses the NuGEN Ovation RRBS Methyl Seq kit
- Complete solution - Enzymatic digestion, library preparation, bisulfite conversion, sequencing and bioinformatics
- Results provided within 6-8 weeks (standard project)

RRBS SERVICE DETAILS

RRBS uses less genomic DNA than WGBS, and reduces sequencing by only requiring coverage of a reduced, representative sample of the whole genome.

- Suitable for genomes with a reference (human, mouse, rat and other mammals).
- Not recommended for plants due to their methylation mechanisms

SUBMISSION DETAILS

- 1 µg of DNA per sample requested (≥20 ng/µl)
- We recommed 100 bp single read sequencing

READS

30M reads (or read pairs) per sample.
Given the nature of the method, variations to this recommendation are possible to meet your experimental needs. We'll be happy to discuss what best suits your aims.

Bioinformatic analysis options are available upon request.

Our funding partners

AGRF is a not-for-profit organisation supported by the Commonwealth Government infrastructure schemes administered through Bioplatforms Australia.

These schemes include NCRIS, EIF, Super Science Initiative CRIS and NCRIS 2.

NCRIS National Research Infrastructure for Australia
An Australian Government Initiative

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