Sequencing of Methylated Human DNA Enriched through MBD-Affinity is a Cost-Effective Alternative to Whole Genome Bisulfite Conversion

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INTRODUCTION

DNA methylation plays a critical role in gene regulation that influences normal organism development and many diseases including cancer. Profiling the DNA methylation patterns of regions of the genome such as promoters, enhancers, or entire chromosomes can provide clues to the functional assessment of transcriptional repression mechanisms and their role in the regulation of gene expression. The methylation of CpG islands (CG-rich regions) has been shown to be associated with segments of the genome that are involved in the development of cancer. However, the analysis of the methylation status of the genome has been made possible by the development of high-throughput sequencing technology and an understanding of how this DNA methylation is controlled. Methylated DNA enrichment and SOLiD™ System sequencing.

MATERIALS AND METHODS

- **Whole-genome SOLiD™ bisulfite sequencing.**
  - Genetic DNA from cultured HFF-10 cells was purified with PureLink columns. Genetic DNA was fragmented to 50-400 bp (mean 250 bp) with a Covarrribi S2 System (Wilmington, MA). Methylated DNA sequencing was performed on the SOLiD™ System using the SOLiD™ D16 System (Life Technologies, Carlsbad, CA) with an average of 8.2× coverage of the methylated genome.

- **RESULTS**
  - **Figure 1.** MethylMiner™ Kit Fractionation of CpG-methylated DNA for SOLiD™ System Sequencing
  - **Figure 2.** Workflow for coupled preparation of unconverted and bisulfite-converted SOLiD™ fragment libraries
  - **Table 1.** SOLID™ System DNA Methylation Analysis Tool
  - **Table 2.** Paired-end mapping state for HFF libraries with and without ECC
  - **Figure 3.** MethylMiner™ Elution profile from 50 μg of human cell line MM-158 genomic DNA (and 500 ng, inset)
  - **Figure 4.** SOLID™ System DNA Methylation Analysis Tool
  - **Figure 5.** Enrichment trends for human genome features
  - **Table 3.** SOLID™ System DNA Methylation Analysis Tool
  - **Figure 6.** MethylMiner™ Elution profile from 50 μg of human cell line MM-158 genomic DNA (and 500 ng, inset)

- **CONCLUSIONS**
  - **Table 3.** SOLI D™ System DNA Methylation Analysis Tool
  - **Table 2.** Paired-end mapping state for HFF libraries with and without ECC

- **REFERENCES**
  - **Table 4.** SOLID™ System DNA Methylation Analysis Tool

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