Locus-specific DNA methylation analysis via amplicon deep sequencing

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Abstract:
Recent advances in sequencing technologies have allowed for the genome-wide quantitative profiling of complete methylomes down to single-base resolution. However, this approach is still both labor and cost intensive and is not readily applicable to focused analyses of particular loci across large sample cohorts. Alternative chip-based technologies either lack the desired resolution or the quantitative readout. Presented here is a novel approach that leverages both PCR amplification as well as deep sequencing of bisulfite-converted DNA to record highly quantitative and locus-specific methylation information for a large number of samples. This involves the automated design of methylation-specific primers that optimally tile the region of interest, alignment of the bisulfite-converted reads, quantification of methylation, and finally the graphical visualization of the results. We have successfully applied this approach in a pilot study aiming to identify novel biomarkers for non-genotoxic carcinogenesis.