

BS-seq reads

Obtain BS-seq reads with replicates for different samples

Quality filter

Filter the reads with FASTQC for bad quality reads

Mapping on genome

Map with bismark tool on genome

Removing PCR duplicates

Remove duplicated reads mapped on bam file

Calling DNA methylation

Call genome-wide DNA methylation for each replicate with bismark

Removing false positive with binomial distribution

Remove false positive experimental error by using discrete probability distribution

Annotating with ChIPseeker: Bioconductor

Assign each methylation site its location on genome and genes

Plotting average profile and heatmap: deeptools

Downstream analysis for visualization

Differential methylation: Per site and per region

Change in methylation among two samples (conditions)