

# THE DIAGENODE EPIGENETICS CUSTOM SERVICE

## Complete workflows for genome-scale DNA methylation and histone marks analysis

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### ABSTRACT

Epigenetics is crucial for the regulation of gene expression and has broad relevance in biological processes like development, disease and response to the environment. For more than 10 years Diagenode has been developing innovative tools to study epigenetic marks such as post-translational modifications of histones and DNA methylation. We are now utilizing our expertise by offering custom services. Our services include full workflows for ChIP-seq as well as reduced representation bisulfite sequencing (RRBS) with our new optimized "Premium RRBS" technology. In addition, we also offer bioinformatic analysis of your results, both standard and customized. The Diagenode Epigenetics Custom Services helps you to complete your epigenetics workflow from your starting biological material to your final results.

### INTRODUCTION

Diagenode offers a wide range of products for every step of your epigenetic analysis. Our chromatin immunoprecipitation (ChIP) solutions are powerful tools to study the association of protein to DNA for analysis of epigenetics modifications, chromatin remodeling and regulation of gene expression by transcription factors. DNA methylation can be studied using reduced representation bisulfite sequencing (RRBS). The Diagenode Premium RRBS™ technology provides high coverage of up to 4 million CpGs for human, distributed mainly in CpG islands and promoter regions, as well as in other genomic elements such as enhancers, CpG island shores and non-coding RNAs.

Diagenode offers ChIP-seq and RRBS as custom services. We process your samples through a complete workflow leading to high-quality analyzed data.

#### Chromatin Immunoprecipitation Sequencing (ChIP-Seq)

##### 1 Workflow

The complete workflow for ChIP-seq contains 6 steps (Figure 1) and results can be provided within 8-10 weeks for a standard project. The service is flexible as researchers may choose either to order the full process or only some specific steps

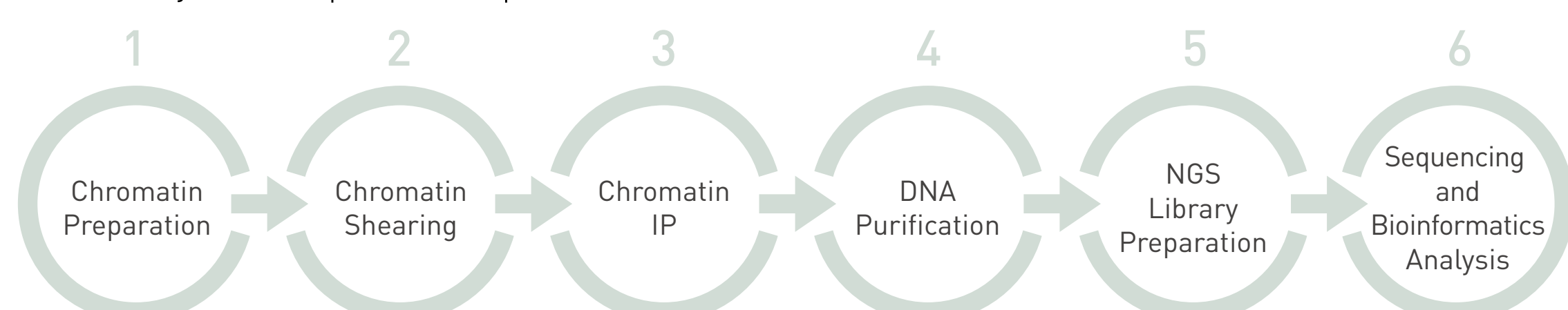


Figure 1. ChIP-seq workflow. Description of the complete procedure for ChIP-seq

Diagenode uses its chromatin shearing and ChIP optimization expertise to find the protocol that is most suited to your model. Moreover, we include several quality control steps to validate the quality of the libraries before Illumina® sequencing.

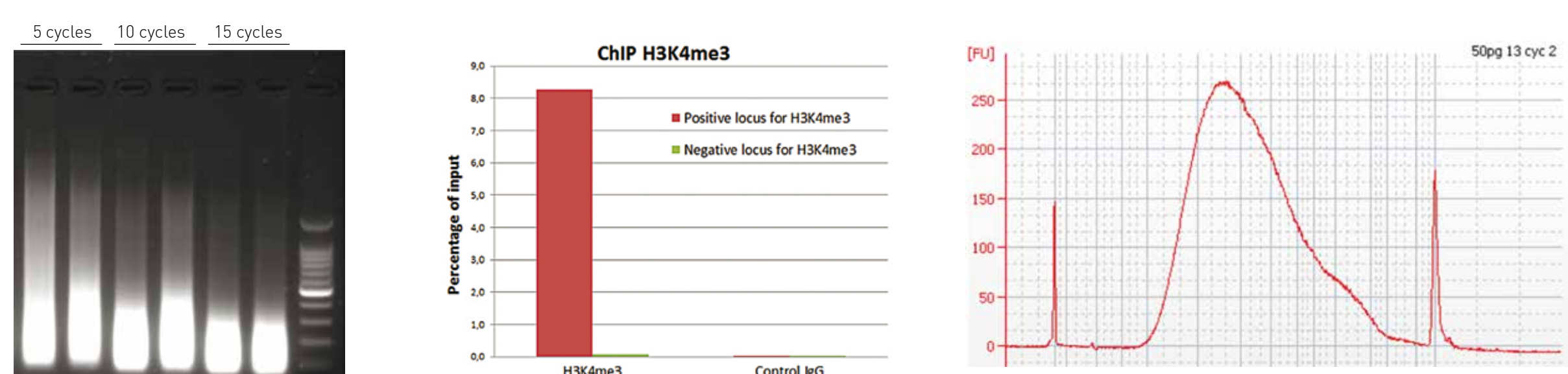


Figure 2. ChIP optimization and quality control. For every new service, several conditions are tested and the efficiency of (a) chromatin shearing, (b) ChIP Immunoprecipitation and (c) library preparation are thoroughly checked.

#### Reduced Representation Bisulfite Sequencing (RRBS)

##### 1 Workflow

The RRBS protocol includes the use of methylated and unmethylated spike-in controls and the multiplexing of the samples. A precise volume is selected during quantitative PCR for each sample prior to pooling to ensure a balanced representation of each sample in the pool.

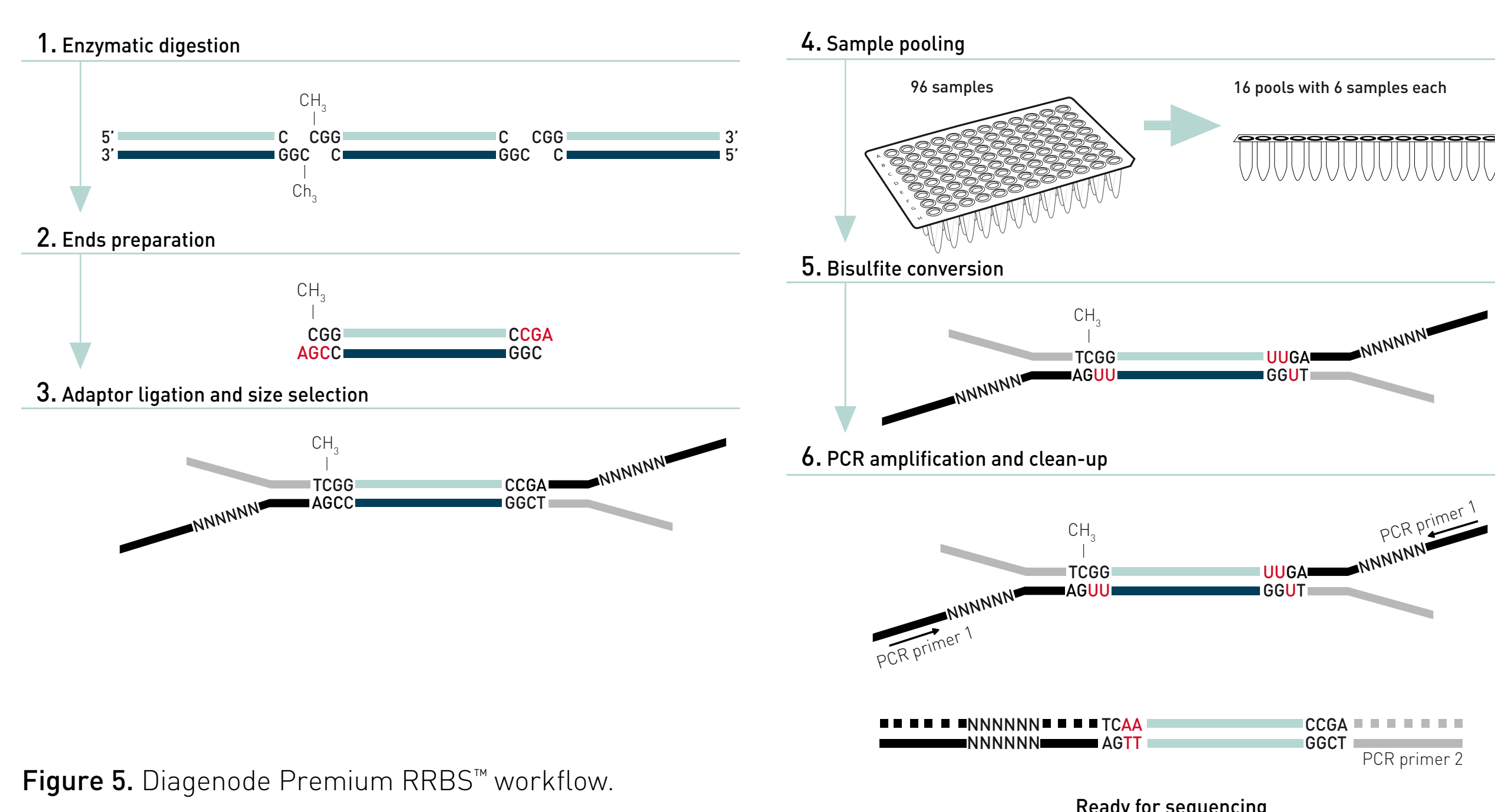


Figure 5. Diagenode Premium RRBS™ workflow.

##### 2 ChIP-seq results

Diagenode's expertise ensures the production of high quality data compliant with existing ENCODE standards.

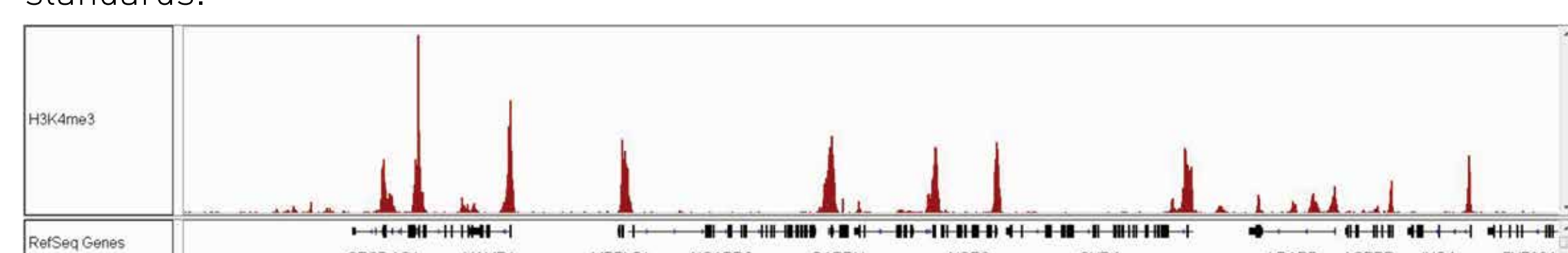


Figure 3. Reads enriched in a genome browser. This above experiment used HeLa cells and the Diagenode Premium H3K4me3 antibody in combination with the iDeal ChIP-seq kit.

Diagenode's expert team of bioinformaticians can perform custom analysis to help you make the most of your data.

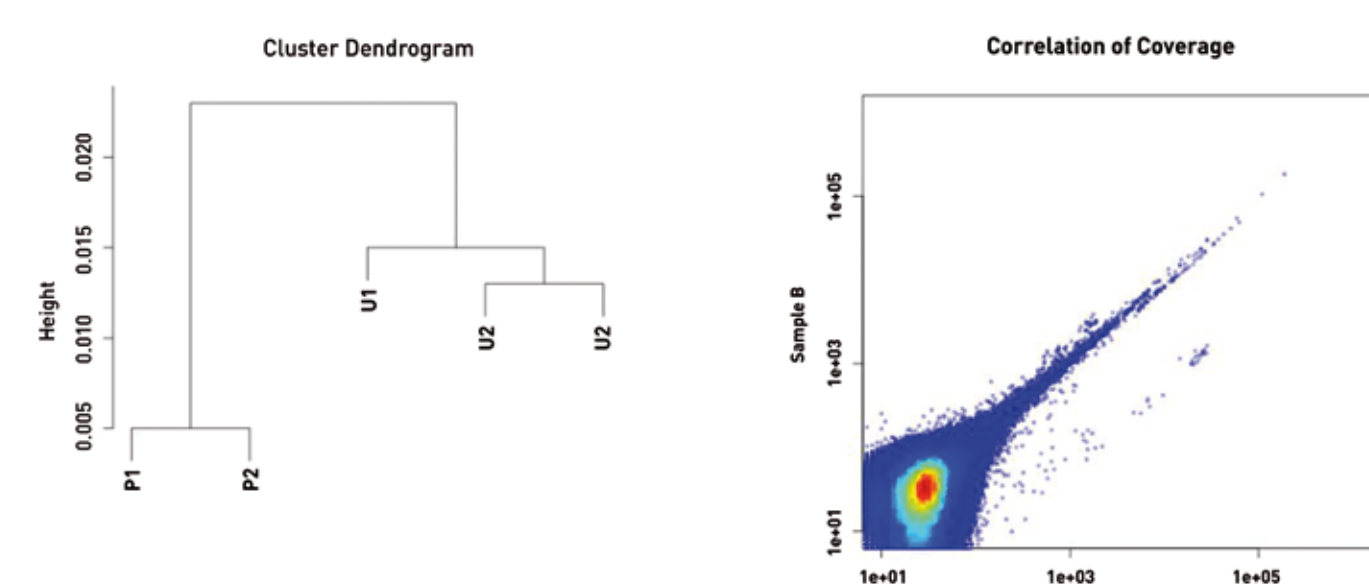


Figure 4. Examples of tailor-made bioinformatic analysis performed on ChIP-Seq data. Many types of analysis can be provided such as hierarchical clustering (a) or correlation curves (b).

### Bioinformatics

Bioinformatics is an integral part of our service with several options available.

Analysis	Features	benefits
Standard	<ul style="list-style-type: none"> <li>Read filtering and trimming</li> <li>Read specific alignment</li> <li>Enrichment specific peak calling/CpG methylation extraction</li> <li>QC metrics</li> </ul>	<ul style="list-style-type: none"> <li>Basis of all analysis</li> <li>Clear and easy to understand</li> <li>Instant evaluation of data set</li> <li>Provides flexibility for your desired downstream analysis</li> </ul>
Comparative	<ul style="list-style-type: none"> <li>Multi-sample cross-comparison</li> <li>Comparison with reference</li> </ul>	<ul style="list-style-type: none"> <li>Multi-sample comparison allows decisive determination of similarities and differences (e.g. between healthy and tumor cells)</li> <li>Best quality assessment using comparisons with highly reliable reference data sets</li> </ul>
Custom	<ul style="list-style-type: none"> <li>Tailored to your project needs</li> <li>Options include                             <ul style="list-style-type: none"> <li>Peak/Methylation profiling</li> <li>Pathway analysis</li> <li>Visualization of regions of interest</li> <li>Annotation</li> <li>Custom graph (publication-ready)...and much more</li> </ul> </li> </ul>	<ul style="list-style-type: none"> <li>Your unique project gets its unique analysis</li> <li>No need for your own bioinformatician</li> <li>Our expert consultation and analysis is customized for your project</li> <li>You name your requirements -- we deliver!</li> </ul>

Table 2. Description of the bioinformatics analysis options

##### 2 RRBS results

RRBS technology can be used for any vertebrate species. The expected library profile depends on the distribution of MspI restriction sites in the genome and is specific to each species.

Species	No. of reads aligned (Alignment rate)	No. of unique CpGs covered	Bisulfite Conversion Rate	Mean coverage per CpG	Library profile
Human	21,787,346 (74%)	3,913,287	99.4%	14	
Human FFPE	27,290,117 (79%)	2,525,053	99.7%	28	
Rat	27,499,313 (86%)	1,663,104	99.0%	38	
Dog	14,053,009 (84%)	3,572,384	99.4%	9	
Zebrafish	29,035,877 (83%)	1,685,466	99.3%	48	

Table 1. Examples of RRBS data generated with the Diagenode Premium RRBS™ technology. Typical data obtained when using human fresh-frozen and FFPE samples, rat, dog, and zebrafish samples.

### CONCLUSION

The Epigenetics Custom Service is now available !

- / Full start-to-finish service - fast and affordable
- / Superior coverage for DNA methylation studies
- / Expertise advantage: 10 years of epigenetics experience; official partner of IHEC-BLUEPRINT Epigenome Consortia
- / Customized support to match your needs
- / Dedicated in-house expert coordinates your project
- / No-obligatory quote request on our website: <https://www.diagenode.com/categories/services>