

# **GUIDELINES**

# ATAC-seq Service Guidelines ATAC-seq 解析サービスガイドライン

ATAC-seq (**A**ssay for **T**ransposase-**A**ccessible **C**hromatin using **seq**uencing) allows for assessing genome-wide chromatin accessibility. The technology is based on the use of the transposase Tn5 which cuts exposed open chromatin and simultaneously ligates adapters for subsequent amplification and sequencing.

ATAC-seq 解析は、transposase Tn5 が露出したオープンクロマチンを切断すると同時に増幅および配列決定のためのアダプターを挿入することで、ゲノムワイドなクロマチンアクセシビリティの評価を可能にします。

In order to maximize results we provide strict guidelines for sample preparation (cell collection). 結果を最大化するため、細胞採取に関するガイドライン(別紙「ATAC-seq 解析 サンプル調製・送付ガイドライン」)を厳守してください。

We request 50.000-100.000 cells / sample to be processed. Cell viability should be higher than 95% when collected.

解析に必要な最低細胞数は1サンプルあたり50,000-100,000個で、回収時の細胞生存率は95%以上です。

# ATAC-seq workflow ATAC-seq 解析内容

Cells will be prepared by the customer according to Diagenode's sample preparation guidelines. Diagenode recommends to perform the experiment at least on two biological replicates.

弊社の細胞サンプル調製手順(別紙「ATAC-seq 解析 サンプル調製・送付ガイドライン」)に従って、細胞サンプルをご準備ください。実験では少なくとも2検体での解析が推奨されます。

#### 1. Sample tagmentation

- Lysis
- TDE1 digestion

#### 2. Library preparation

- Library amplification
- Library purification
- QC on the ATAC-seq library (DNA concentration, analysis of library profile)
- Libraries pooling

# 3. Deep sequencing

- Samples are sequenced on an Illumina ® platform, paired-end 2x75bp
- 100 million raw reads on average are obtained per sample when pooling 3samples/lane (Hi-seq 4000). Sequencing depth will be adjusted to project dependent criteria.

#### 4. Bioinformatic analysis

Quality check, alignment to reference genome, identification of enriched regions (peak calling) are included.



# **GUIDELINES**

# Provided files:

### 納品データ

- Report with sequencing statistics
- Raw data in FASTQ format
- FastQC reports
- Alignment files in BAM format
- O Peak files in BED format

### Additional analysis on request:

# 追加解析オプション

• **Differential binding analysis:** Identification and annotation (human, mouse, rat, drosophila) of differential chromatin accessibility between samples.

#### **Provided files:**

- O Report with summary of differential accessibility analysis and plots
- Files containing differentially accessibility sites or unique peaks and breakdown of those in annotated regions: introns, exons, promoters, 1-to-5 kb upstream-TSS and intergenic regions for human, mouse, rat and drosophila.
- Annotation in genomic regions: Annotation of ATAC-Seq peaks with genomic regions: introns, exons, promoters, 1-to-5 kb upstream-TSS and intergenic regions for human, mouse, rat and drosophila.
- Gene ontology terms analysis: Enrichment analysis on gene sets. Gene Ontology terms that are
  overrepresented in open chromatin regions or differentially accessible regions may indicate the underlying
  biological processes involved.
- Pathway analysis: Identify biochemical pathways in which genes associated with open chromatin regions
  or differentially accessible regions may be overrepresented.
- **Visualization of specific genomic regions:** Visualization of results (i.e. sequencing data, peaks) at specific genomic regions (e.g. genes, promoters) in publication-ready images (human, mouse, rat).



# **GUIDELINES**

# Additional information その他の注意事項

For sample preparation and sample shipment it is mandatory to follow Diagenode's guidelines. If customer samples do not meet Diagenode's quality requirements, any additional QC of new samples will be charged to the customer. Any delay in sample shipment to Diagenode's facilities might result in delaying customer's project. サンプル調製・送付については必ず弊社のガイドライン(別紙「ATAC-seq 解析 サンプル調製・送付ガイドライン」)に従ってください。ご提供いただいたサンプルの品質が基準に満たない場合、再度新しいサンプルをご提供いただき追加の品質確認を行いますが、再提供および品質確認にかかる追加料金はお客様負担となります。弊社施設へのサンプル提供の遅れは、納期に影響する可能性がございます。

Generated files will be available for download during 1 month and stored for an additional period of 3 months on Diagenode's servers. Additional long-term storage of data is available upon request. This offer includes a one hour call to walk you through the results if needed.

解析結果データは弊社 WEB サイトよりダウンロード可能です。解析終了後、Diagenode SA (ベルギー) よりログイン及びダウンロード方法についてご連絡いたしますので、1ヶ月以内にダウンロードをお願い いたします。また、予備期間として 3 か月間はサーバー上に保存されていますが、それ以降は削除され ますので、長期間の保存を希望される場合は早めにご連絡ください。必要であれば解析結果について Diagenode SA(ベルギー) の担当者と 1 時間通話することもできます(アポイントメント後、英語でのご説 明となります)。

Original samples are stored at Diagenode during 12 months after project completion, but will be discarded once this time is exceeded. Return shipment of samples is available upon request.

お客様からお預かりしたサンプルは弊社施設にて解析終了から 12 か月間保管した後、順次廃棄いたします。お客様のご要望に応じて、サンプルのご返却も可能です(追加費用がかかります)。

Any additional service which is beyond the current project scope will be charged to the customer. ご依頼いただいた内容の範囲外となる追加サービスの料金はお客様負担となります。