



Assay for Transposase-Accessible Chromatin (ATAC-Seq) data processing workflow

The paired end Illumina ATAC-Seq raw data was processed using in house scripting (bash and R) on University of Edinburgh high performance computing facility (Eddie3). The ATAC-Seq data statistical analysis was carried out in **R v ≥ 3.6** ¹ using **ChIPseeker v1.26.0**² package for annotation and metrics. Briefly quality control of the raw sequence data was performed using **FastQC v0.11.9**³ and **multiQC v1.9**⁴ (Ewels et al., 2016) programmes. The paired end reads were trimmed using **Trimmomatic v0.39**⁵ (Bolger et al., 2014) and the following trimming criteria:

```
trimmomatic-0.39.jar ILLUMINACLIP:/Trimmomatic-0.39/adapters/NexteraPE-PE.fa:2:30:10:1:true SLIDINGWINDOW:5:20 MINLEN:30.
```

The trimmed reads were then mapped to the Sscrofa11.1 pig reference genome available from Ensembl v100 (GCA_000003025.6) using **Bowtie2 v2.3.5.1**^{6,7} and the default flags of the *--very-sensitive* mode. The BAM files that were generated were then sorted and indexed using **samtools v1.6**⁸. ATAC-Seq peak calling was performed using **Genrich v0.5**⁹ (John M. Gaspar, 2020) under ATAC-Seq mode using the following flags:

```
Genrich -t input.bam -o output_peak.bed -f output_peak.log -z -j -r -e MT -v.
```

Intersectionality of the peaks called was explored using **bedtools' v2.26.0**¹⁰ intersect and subtract functions:

```
bedtools intersect -sortout -u -a A.bed -b B.bed C.bed D.bed > ABCD.bed
```

```
bedtools subtract -A -a A.bed -b ABCD.bed > A_specific.bed
```

References:

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