

Practical S10.1: Mapping PE reads with BWA mem

This practical session is a hands-on exercise to carry out short-read mappings against a reference genome. We will use the most popular read aligner BWA with its BWA-MEM algorithm.

Set up for the practicals:

- Create a new conda environment: `conda create -n read_alignment`
- Activate the environment and install the following packages:
 - `conda install -c bioconda minimap2`
 - `conda install -c bioconda bwa`
 - `conda install -c bioconda samtools`
 - `conda install -c bioconda bedtools`
- Download all the reads and the reference from Aula ESCI
- Install IGV Desktop app: <https://software.broadinstitute.org/software/igv/download>

Mapping PE reads

1. Align the lynx paired-end (PE) reads to the reference mitochondrial genome using BWA-MEM. Manual: <https://bio-bwa.sourceforge.net/bwa.shtml>
2. Convert the alignment into a BAM file (you'll need samtools).
3. Use samtools to obtain mapping stats. How many reads have mapped and how?
4. Use bedtools to estimate coverage
5. Visualize the BAM using IGV.
6. Look at the alignments.
7. How is the coverage along the genome?
8. Is there any region with really high or low coverage? Where it is located?

Mapping Unknown reads

1. Align `unknown_reads.fastq` to the same reference mitochondrial genome using BWA-MEM.
2. From the SAM/BAM file can you tell how many reads are unmapped and how many mapped?
3. Visualize this alignment together with the previous PE alignment on the lynx mitochondrion. Where does this fall and what is the coverage.

Useful Links:

SAM Flags explained: <https://broadinstitute.github.io/picard/explain-flags.html>

Samtools: <https://samtools.sourceforge.net/>

Bedtools: <https://bedtools.readthedocs.io/en/latest/>