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Mapping ONT Reads to Reference Sequences with Minimap2 and Samtools

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Hung Luong[1], Hiep Vu[1]

[1]University of Nebraska-Lincoln

Hung Luong
University of Nebraska-Lincoln

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Protocol status: Working
We use this protocol and it's working

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Abstract

This simple protocol maps ONT raw reads to Reference Sequences by Minimap2 and Samtools.

Install requirement software

1 git clone https://github.com/lh3/minimap2
cd minimap2 && make

2 conda create -n samtools
conda activate samtools
conda config --add channels bioconda
conda config --add channels conda-forge
conda install -c bioconda samtools

Align reads to reference sequences (two options as bellow)

3 # Without index reference sequence
minimap2 -ax map-ont /path/to/list/references.fasta /path/to/raw/reads.fastq.gz > /path/to/output/folder/alignment_reads.sam

4 # Index reference sequences first
minimap2 -d /path/to/index/references.mmi /path/to/reference/sequences/references.fasta
Align reads to reference sequences with ONT reads
minimap2 -ax map-ont /path/to/list/references.fasta /path/to/raw/reads.fastq.gz > /path/to/output/folder/alignment_reads.sam

Keep only mapped reads

5 samtools view -bS -F 4 /path/to/output/folder/alignment_reads.sam > /path/to/mapped/reads.bam

Sort out the mapped reads

6 samtools sort /path/to/mapped/reads.bam -o /path/to/mapped/reads_sort.bam

Convert mapped reads to FASTQ or FASTA file for downstream analysis

7 samtools fastq /path/to/mapped/reads_sort.bam > /path/to/mapped/reads_sort.fastq