



RNA-sequencing Experimental Design and Analysis Methods *CB321*

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<https://hbctraining.github.io/RNA-seq-CB321qc>



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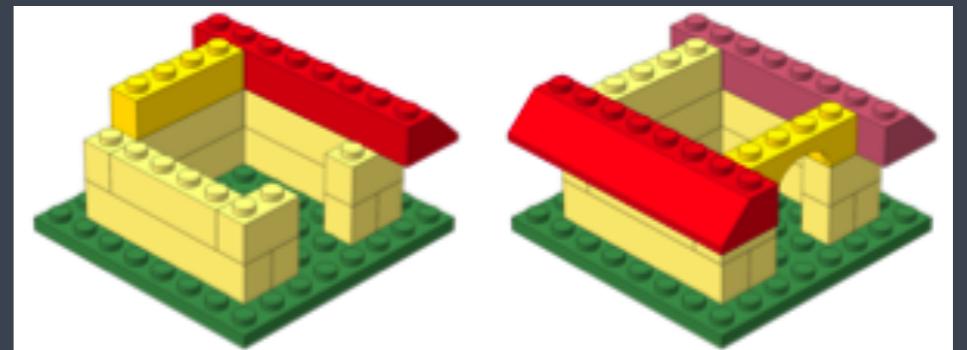


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Transcriptomics (RNA-seq)

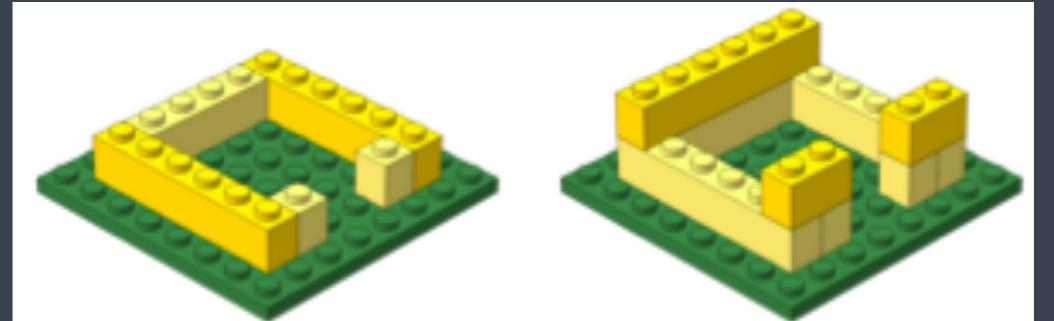
- RNA-seq is the process of sequencing the “transcriptome”
- Uses include:
 - Differential Gene Expression
 - Transcriptional profiling with single cells
 - Transcriptome assembly
 - Metatranscriptomics or community transcriptome analysis
 - Mechanisms of transcription looking at nascent transcripts
- Specific use of RNA-seq will determine experimental protocols, sequencing strategies, and analysis methods

Learning Objectives



- ✓ Describe sequencing by synthesis, including cDNA library preparation.
- ✓ Describe the design considerations to perform an **RNA sequencing experiment**.
- ✓ Describe the workflow for a standard bulk or single-cell RNA-seq data analysis.
- ✓ List computational skills necessary to implement the various steps in the analysis workflow.
- ✓ List genomics-specific databases for data sharing and download

Base components



✓ Unix/Linux shell

- Dealing with large data files (e.g. FASTQ, BAM)
- Using bioinformatics tools (e.g. Salmon, Trinity)
- Accessing and using high-performance compute clusters (O2)

✓ R

- Parsing and working with smaller results files
- Statistical analysis, e.g. differential expression analysis (DESeq2) or single-cell analysis (Seurat)
- Generating publication-quality figures from complex data (ggplot2)

Link for class today

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