Everything you wanted to know about RNAseq

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Overview

Today's Goals

- How does RNAseq work?
- What are the benefits?
- What are the challenges?

- Meet your group!

Dotstorming

- What questions did you have?
- Dotstorming Board
- Write your question
- Vote on others
- (Take 5 mins)

A Parable

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- They decide to grow the same vine in 6 different pots by the window in their classroom

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- What is wrong with this “experiment”?

Central Dogma

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- Protien

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- A written record of selection over time
  - (or population size changes)
  - (or species histories)

RNA

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- Only exons, introns are spliced out

Protein

- Final product of central dogma

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- “Action item” from the list

Protein

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- “Action item” from the list
- Performs a task within the cell

RNAseq

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- Why not DNA?
- Why not Proteins?

Measuring RNA

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3. (Next-Gen) Sequence!!!

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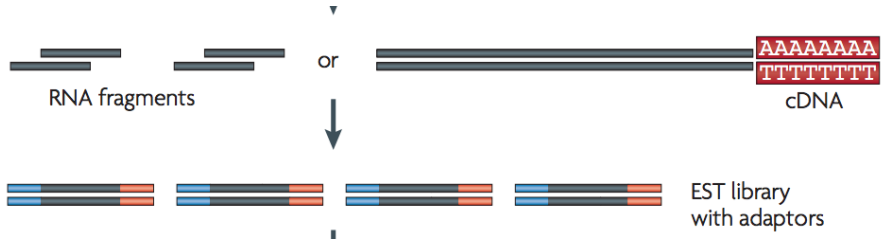
  - Can be kept with labeling

prepare a sequencing library

- Take the now-cDNA into a standard library prep



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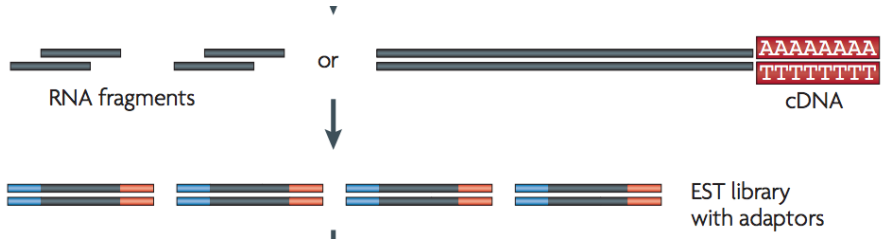
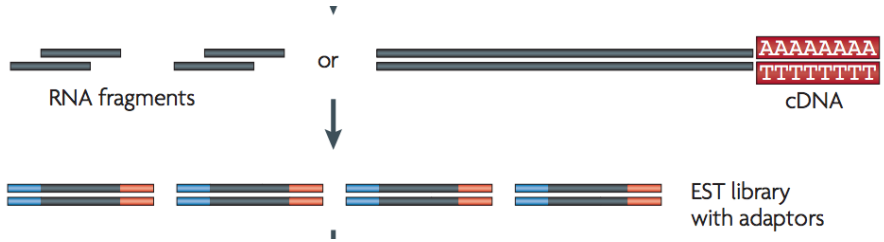


prepare a sequencing library

- Take the now-cDNA into a standard library prep
- Add adapters
- Amplify (with PCR) and sequence



NGS!!!

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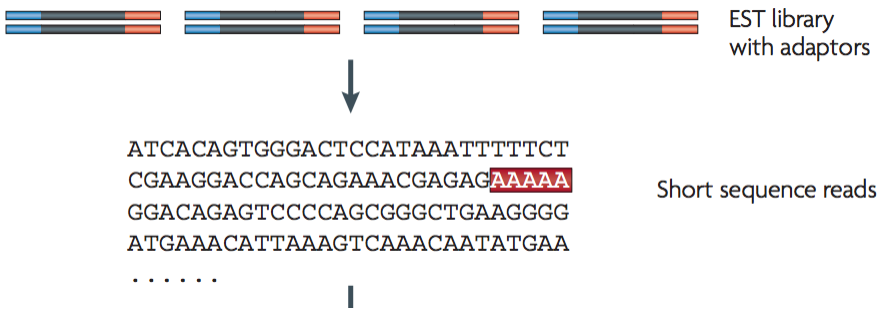

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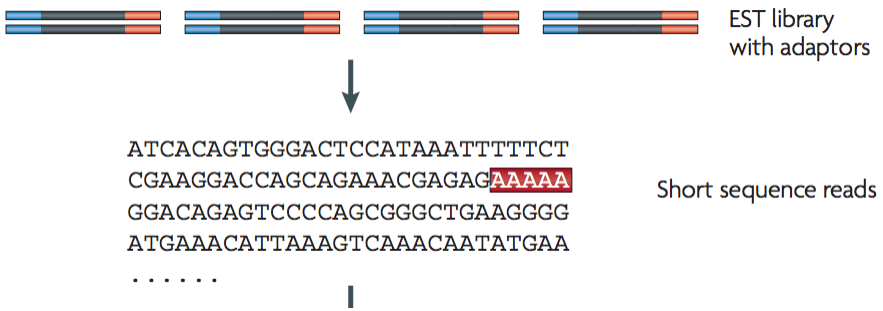

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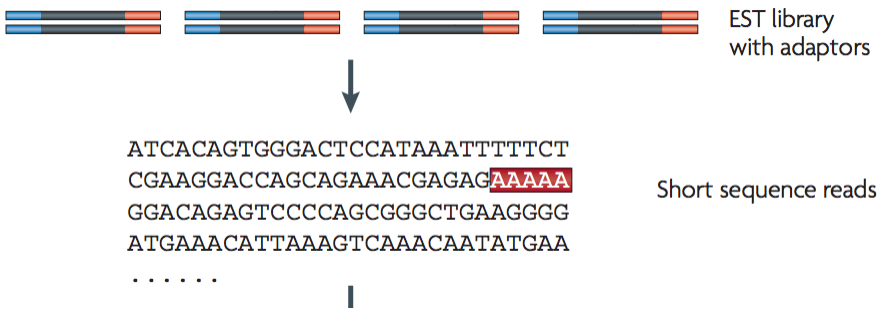

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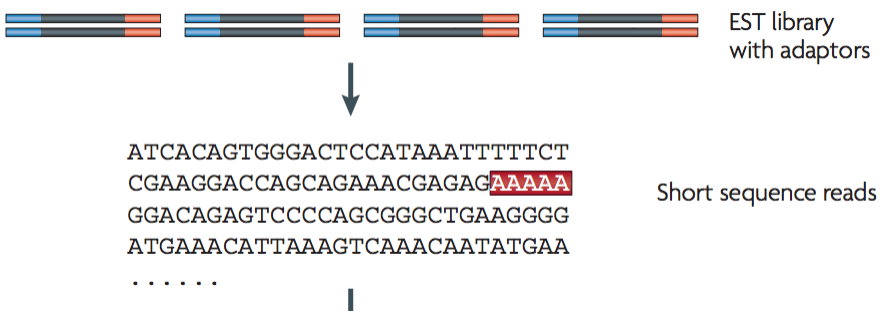

fastq Format

1. Sequence header - information regarding the machine

```
@HWI-EAS209_0006_FC706VJ:5:58:5894:21141#ATCACG/1
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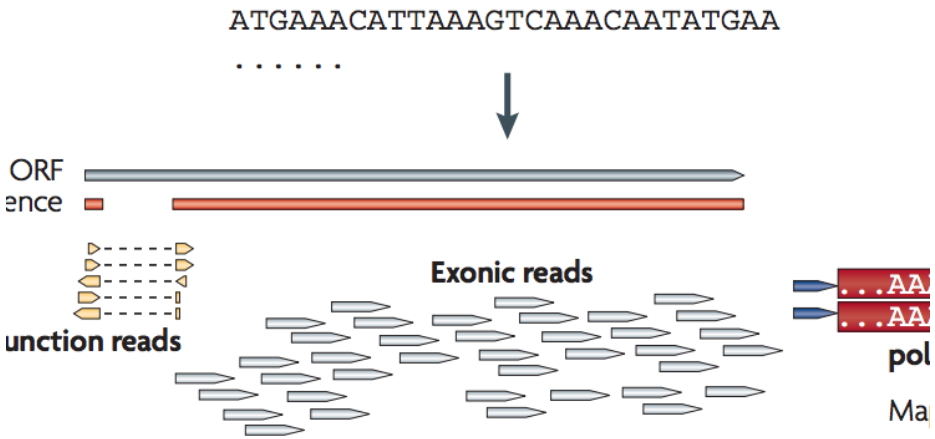

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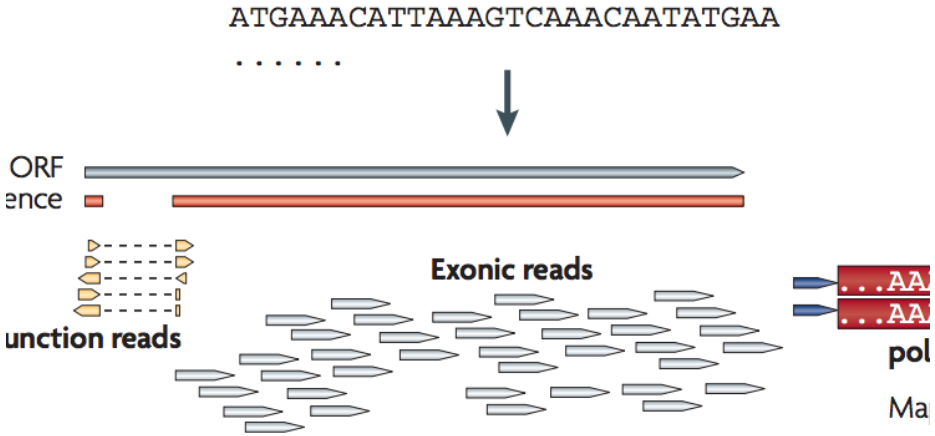

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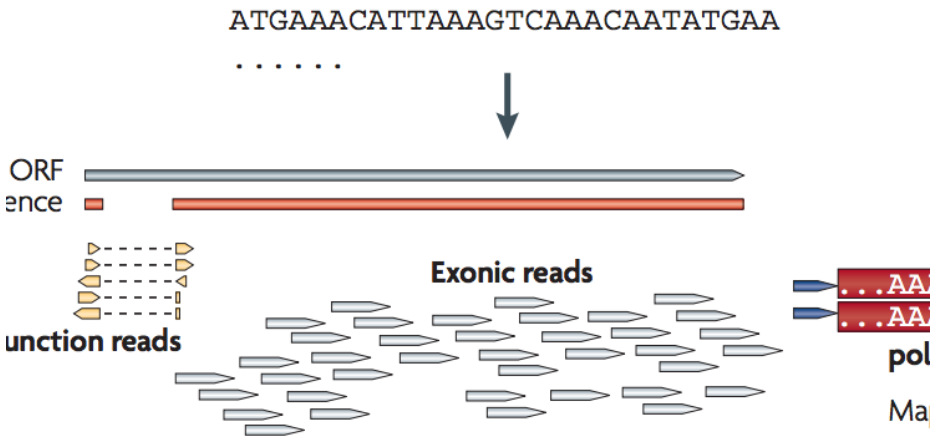

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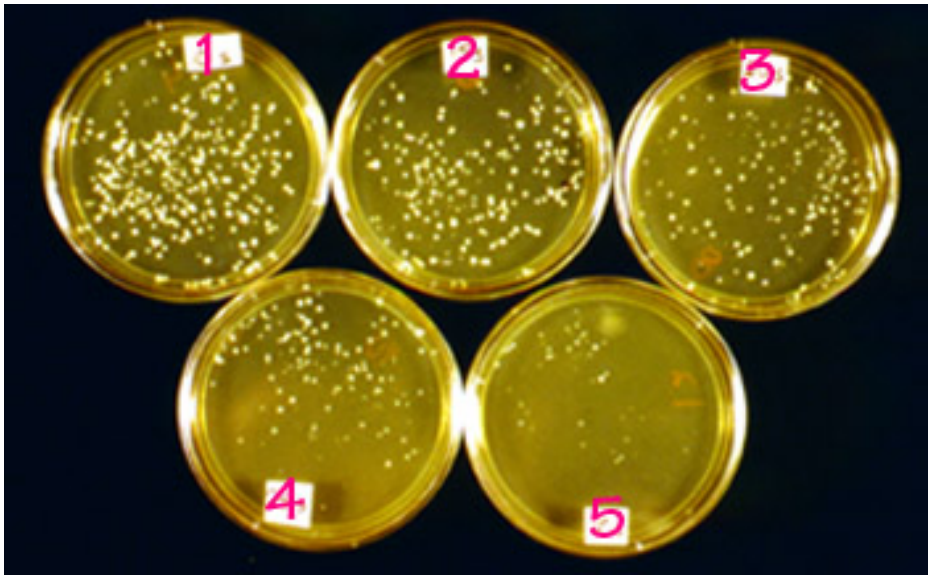

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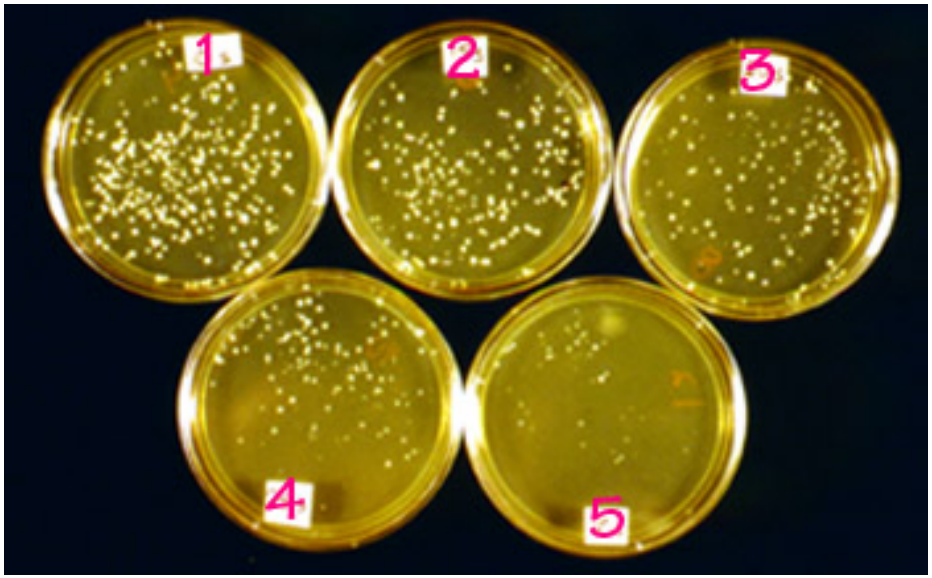
- Steve and Susan wanted to know how food source affects yeast
- They decide to grow the same yeast in 5 different plates in their classroom



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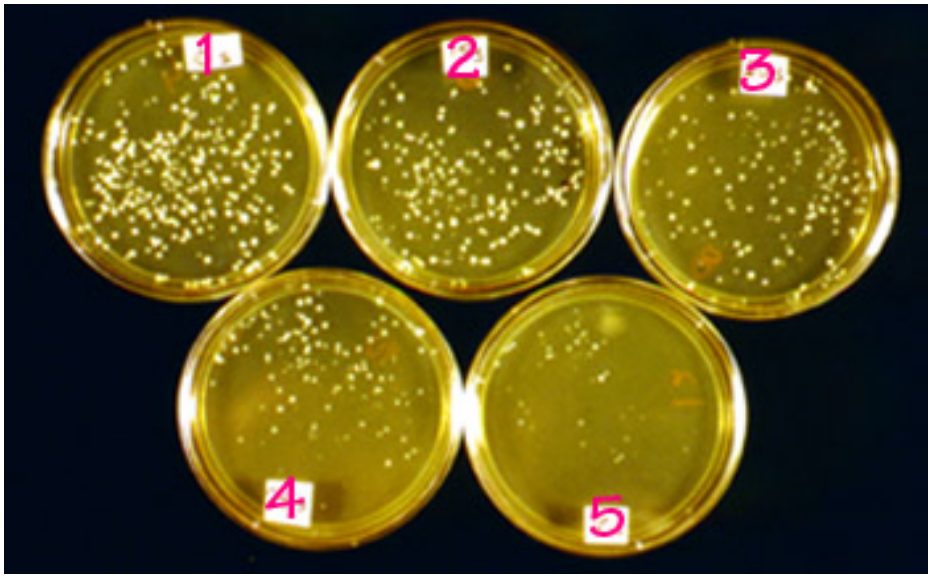

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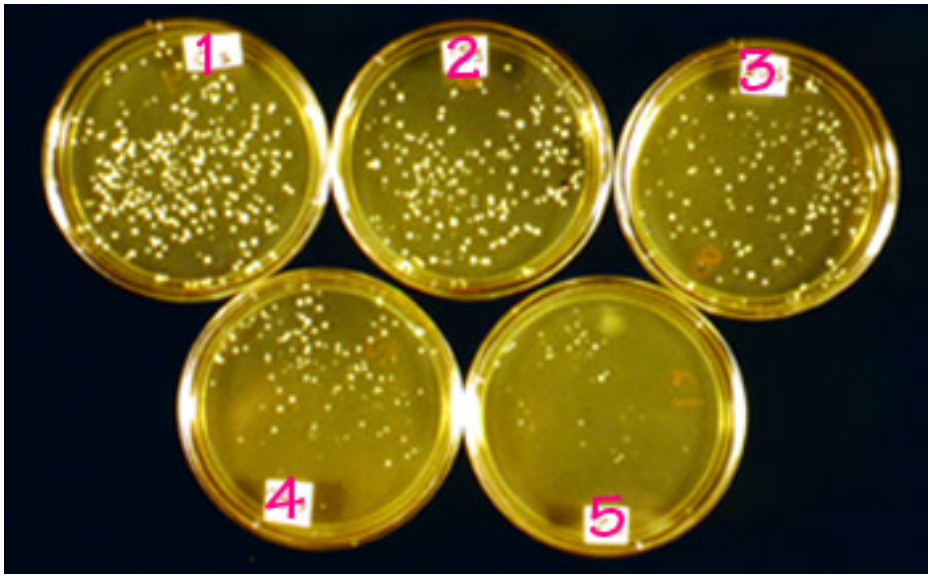

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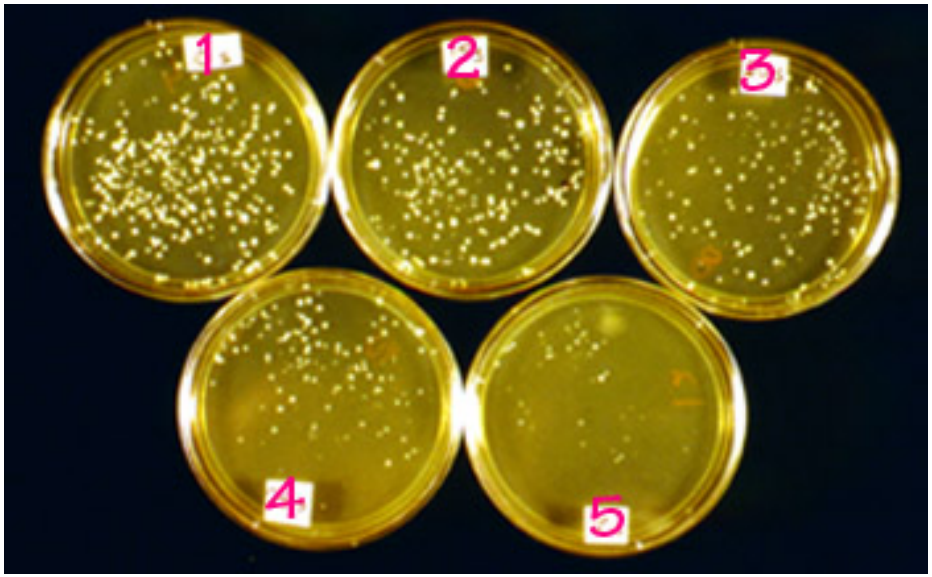

Dotstorming Check-In

- Did I cover the top questions?
- Dotstorming Board

Learning to Code (and other skills)

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- Leave with a plan to find data by next Tuesday

The End