Pathogen Genomics – working with pathogen genomes

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Wellcome Sanger Institute

LSHTM Pathogen Genomics 2020

Summary

- What is the point of a genome sequence?
- Genome sequencing technologies
- Sequence data files
- Viewing genomes

Why do genome sequencing?

- Reference for molecular biology
  - *Tropheryma whipplei* causes the potentially fatal Whipple's disease. Could not easily be grown. Genome revealed it had lost genes involved in producing amino acids.
- Identify all the genes that determine the function of the organism
  - *Neisseria meningitidis*, a major cause of meningitis. The first vaccine for a particular form of meningitis for identified by looking for candidates in its genome.
  - *Rickettsia prowazekii* is the cause of epidemic typhus, which killed millions in the early 20th century. It cannot reproduce outside of these cells. It was found to have just over 800 genes.
- Examine evolution by comparative genomics
- Track spread of pathogens
- Identify antimicrobial/drug resistance genes and drug targets
  - Mtb researchers made bacteria resistant to a new drug. Genome sequencing identified the gene involved in resistance.
- Basis for other omics technologies – RNA-seq, ChIP-seq, Methylome etc.

Why do genome sequencing? - Video of Wellcome Sanger Institute researchers

Technology overview

- Sanger sequencing produces ~500bp reads
  - Pros: Highly accurate
  - Cons: Expensive, laborious
  - Uses: High quality reference genomes
- Illumina's sequencing-by-synthesis 75-250bp
  - Pros: cheap, lots of reads (e.g. 500 million per run)
  - Cons: short reads
  - Uses: Resequencing, draft genomes, RNA-seq
- Pacific Biosciences Single-Molecule Real Time (SMRT) reads of 5000bp-40000bp
  - Pros: long reads
  - Cons: Fewer reads than Illumina - ~1 million, low accuracy
  - Uses: Reference genomes

Genome sequencing technologies – Interview with Mike Quail

Sequence data

Fasta

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RP   1-591
RX   MEDLINE; 99369842.
RA   Brinkman F.S., Schoofs G., Hancock R.E., De Mot R.;
RT   "Influence of a putative ECF sigma factor on expression of the major outer
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RT   fluorescens";
RL   J. Bacteriol. 181(16):4746-4754(1999).
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EMBL Flat File

Header
- Title
- Taxonomy
- Citation

Features (AA seq)

DNA Sequence

SAM/BAM

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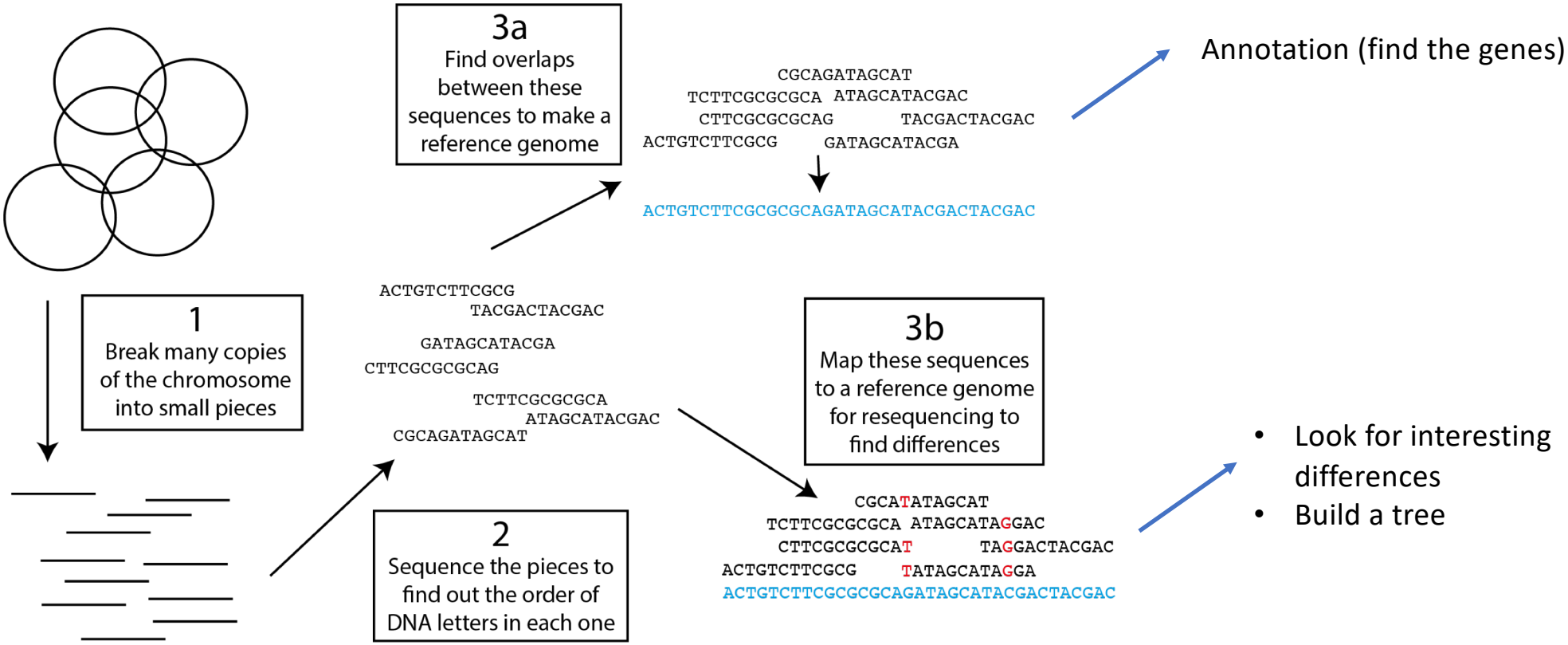
What do we do with these data?



Doing bioinformatics

- Has anyone here done any bioinformatics?
- How are we going to do our bioinformatics?
  - Virtual machine with Linux
  - Artemis for viewing genomes
  - Various command line tools for mapping, assembling etc.
  - Web-based applications

What will we do today?

- Get familiar with the Virtual Machine
- Use Artemis to get intimate with genomes (morning)
- Map Illumina genome sequencing data to understand differences between closely related bacteria (afternoon)