Introduction of Next-Generation Sequencing

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Content

1. Human Genome Project (HGP)
2. Next-Generation Sequencing (NGS)
3. Short read vs Long read
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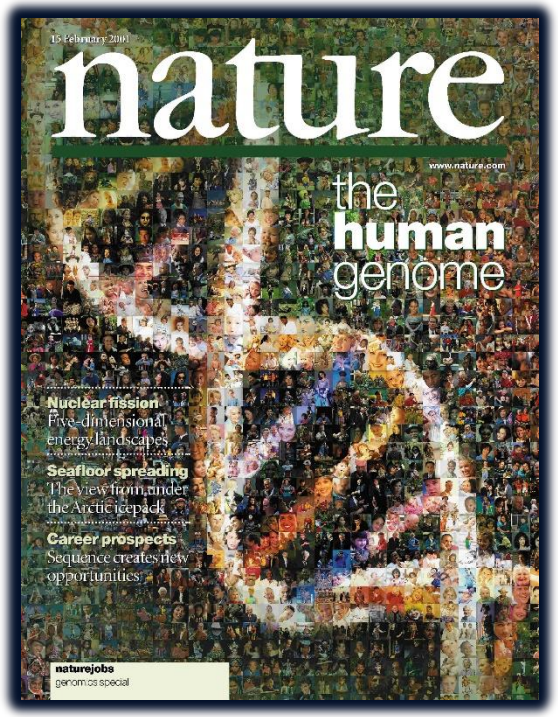
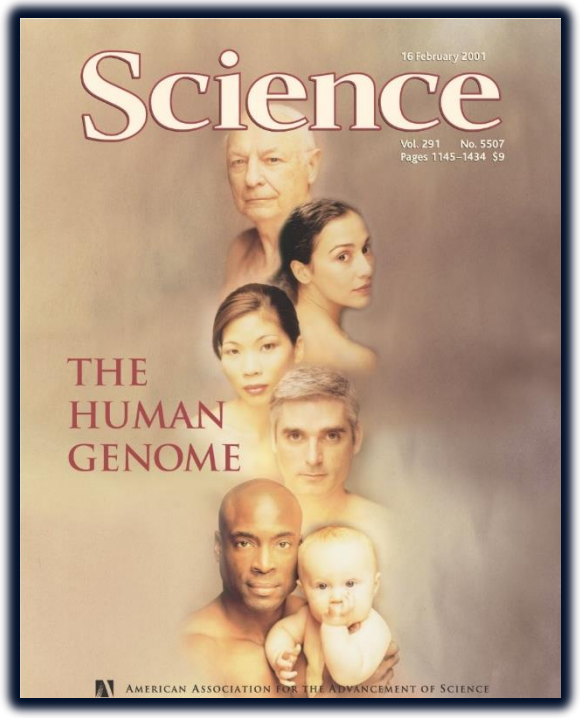
Human Genome Project - HGP (Oct 1990 - April 2003)

1. In 2003, the Human Genome Project produced a genome sequence that accounted for over 90% of the human genome (**~3 GB**).
2. It was as close to complete as the technologies for sequencing DNA allowed at the time.
3. Cost ~3 billion US$

=> Facilitating advancements in **next-generation sequencing (NGS)** technologies



HGP Paper



Venter/Celera Paper

https://www.genome.gov/human-genome-project

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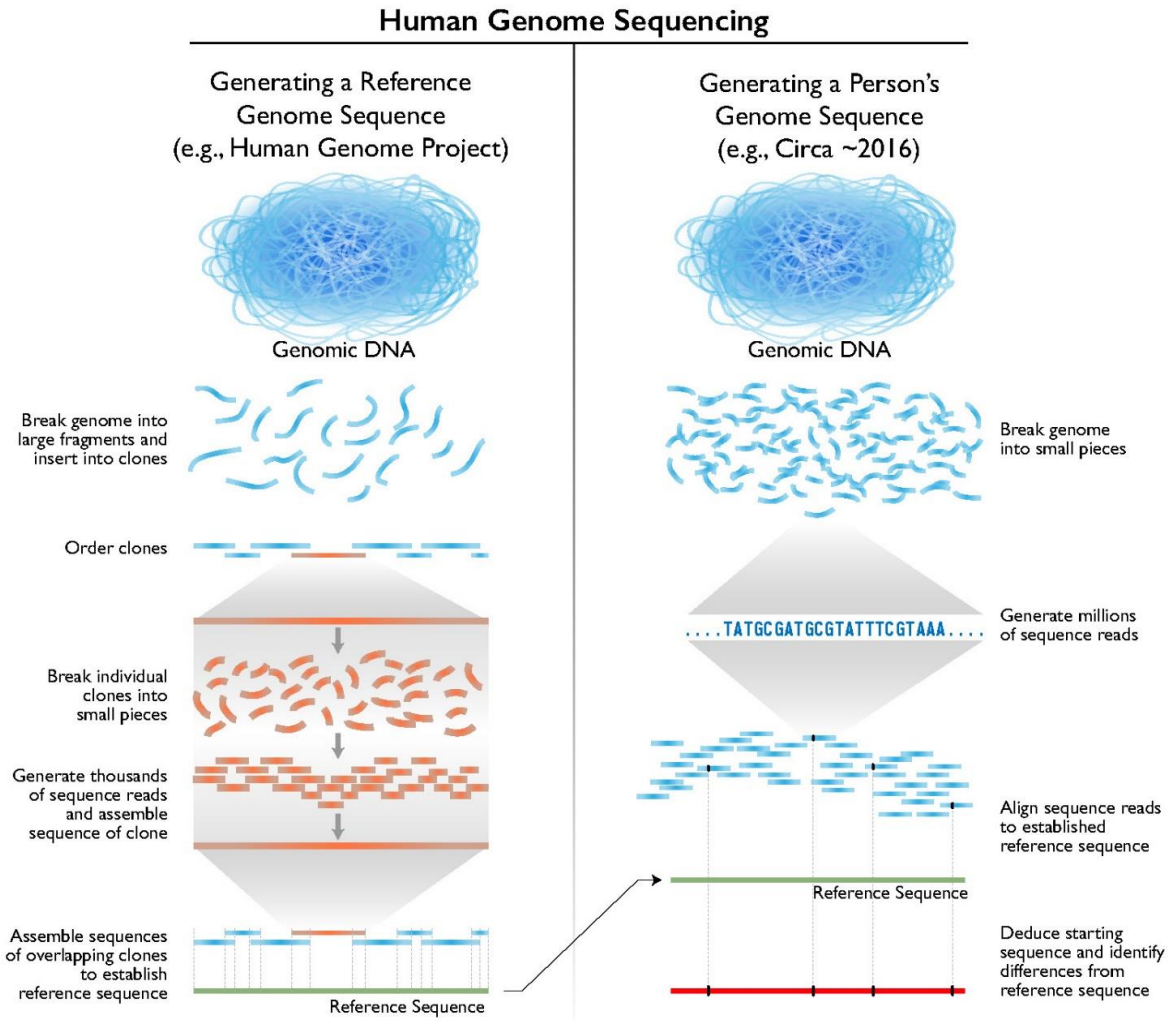
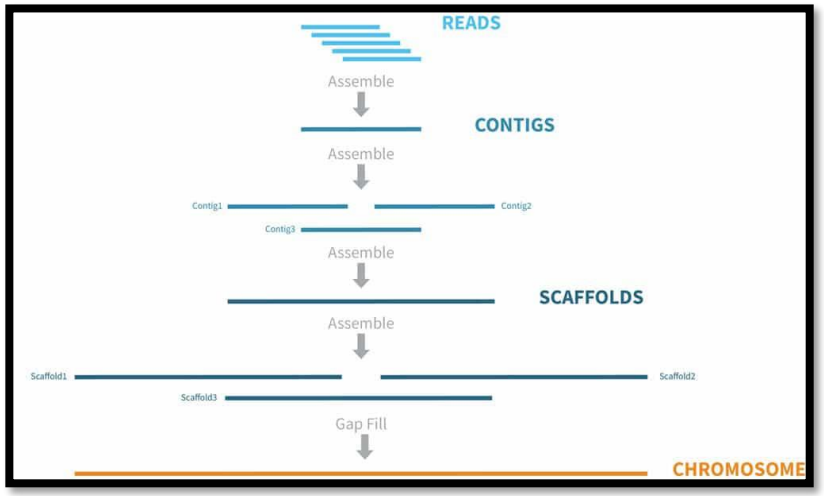
https://www.ncbi.nlm.nih.gov/nuccore/806904736

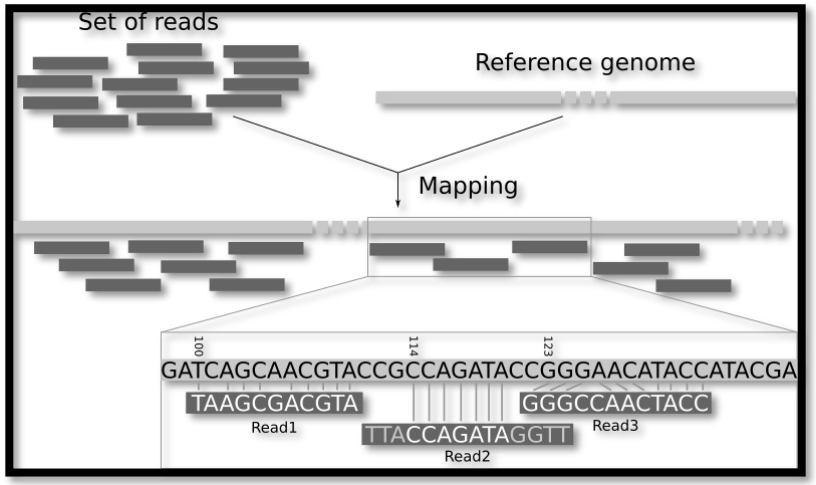
Next-Generation Sequencing (NGS): Reference Genome



De novo assembly

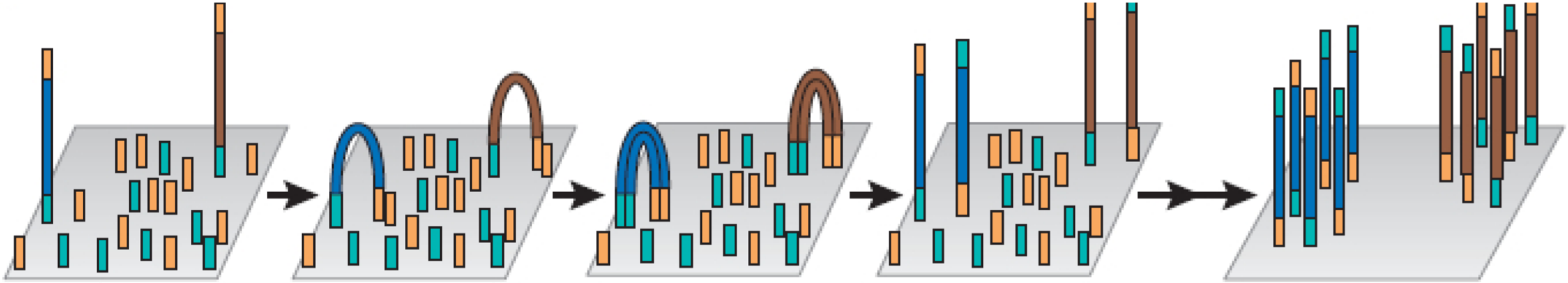


Mapping to reference



https://www.genome.gov/about-genomics/fact-sheets/Sequencing-Human-Genome-cost

NGS: 1) Parallel Sequencing



10.3390/medsci2020098

NGS: 2) Reference Genome

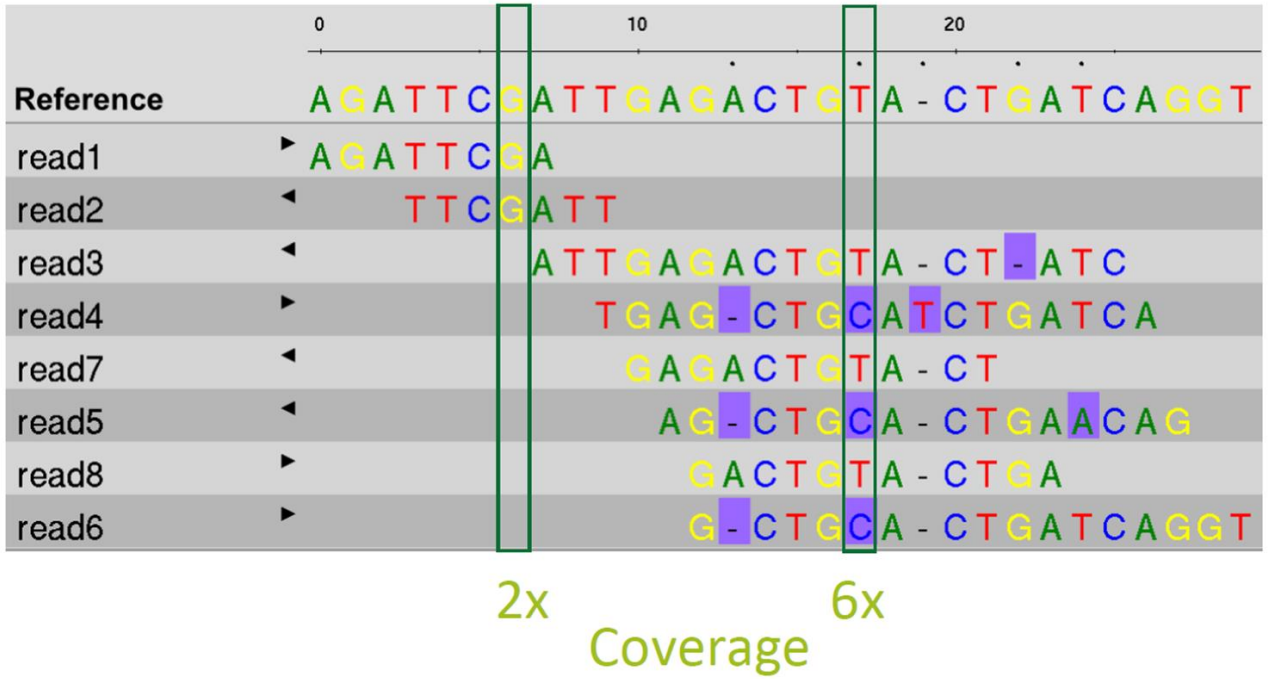
De novo assembly

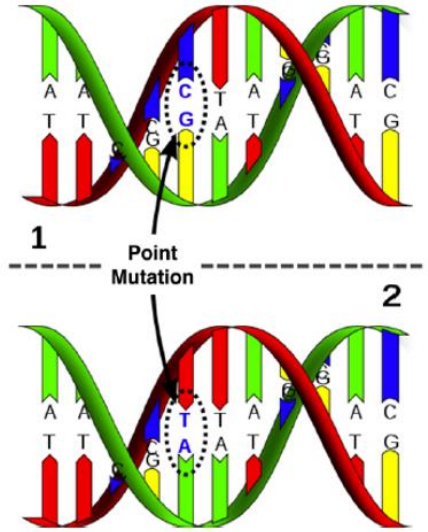
Mapping to reference

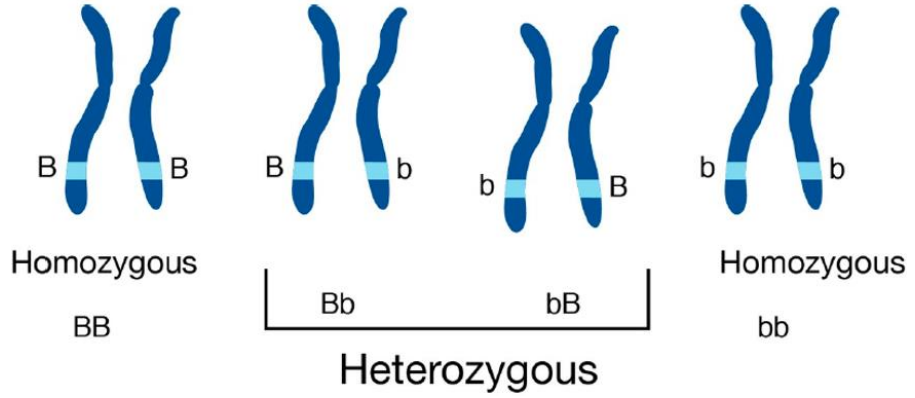
https://www.genome.gov/about-genomics/fact-sheets/Sequencing-Human-Genome-cost

Human Genome Variation

- Human genomes are > 99% similar by sequence
- A typical human genome has ~ 5 million variants with 3-4 million single nucleotide variants
- Humans are diploid

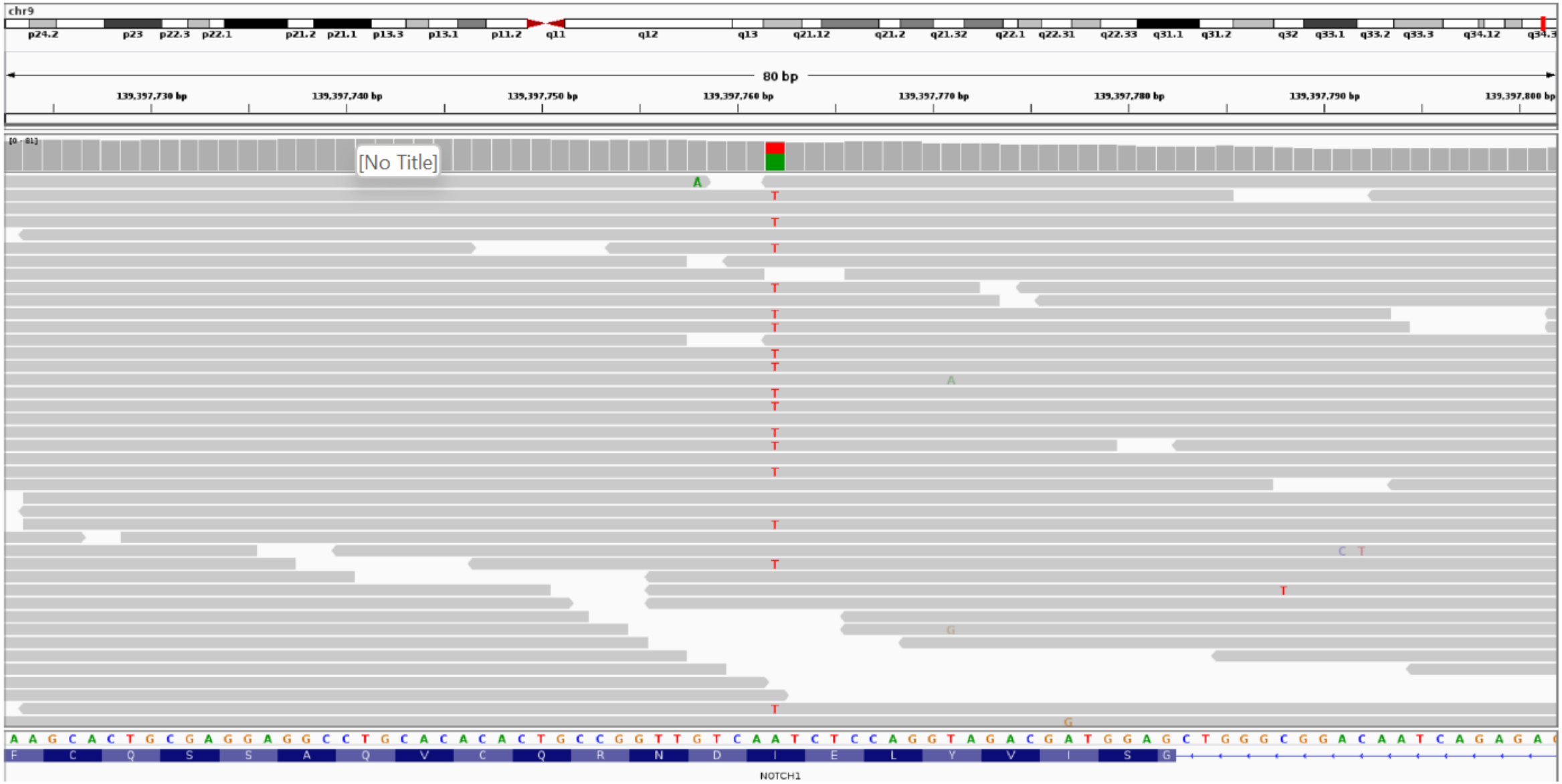




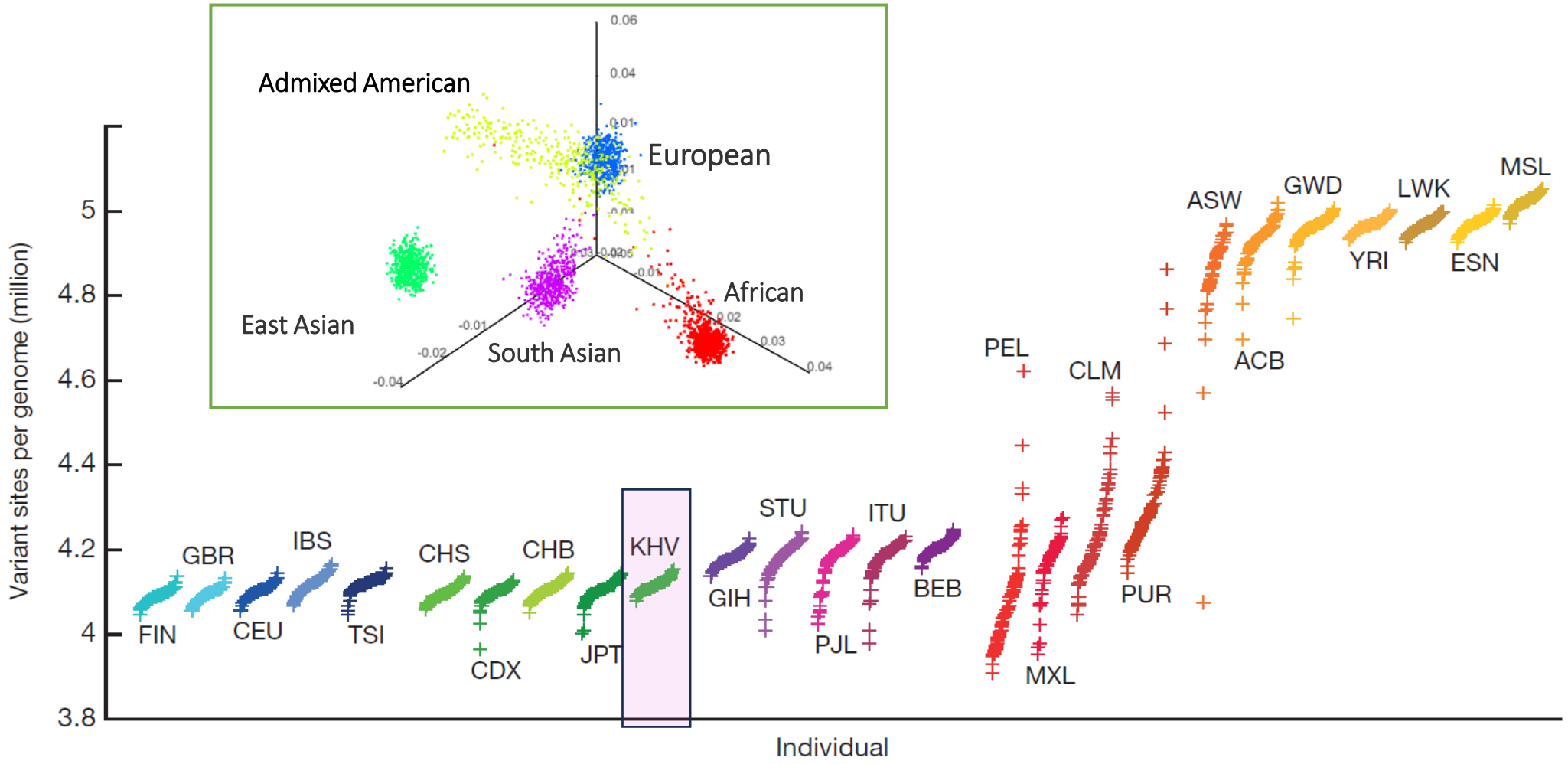


https://rosalind.info/media/point_mutation.png, https://en.wikipedia.org/wiki/Zygosity

Alignment and variant viewers



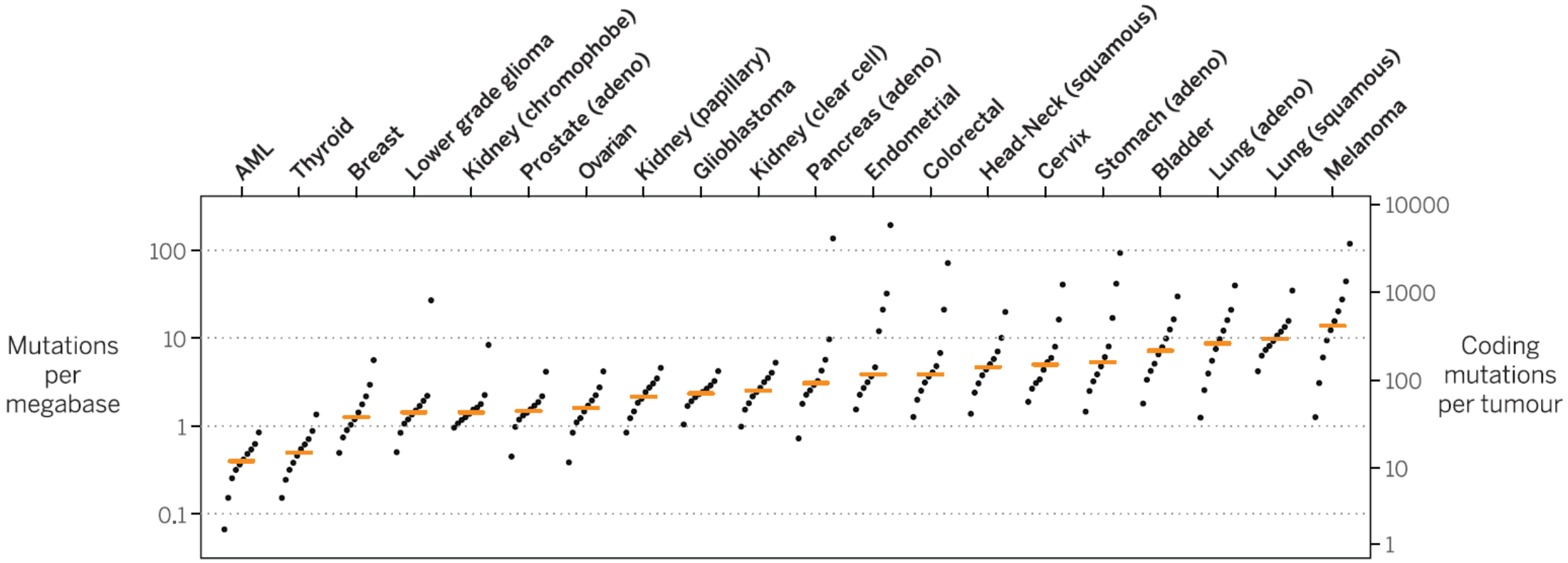
Human Genome Variation: 1000 Genomes Project



The number of variant sites per genome of 1K human genomes project (2015)
Kinh in Ho Chi Minh City, Vietnam (KHV)

doi:10.1038/nature15393

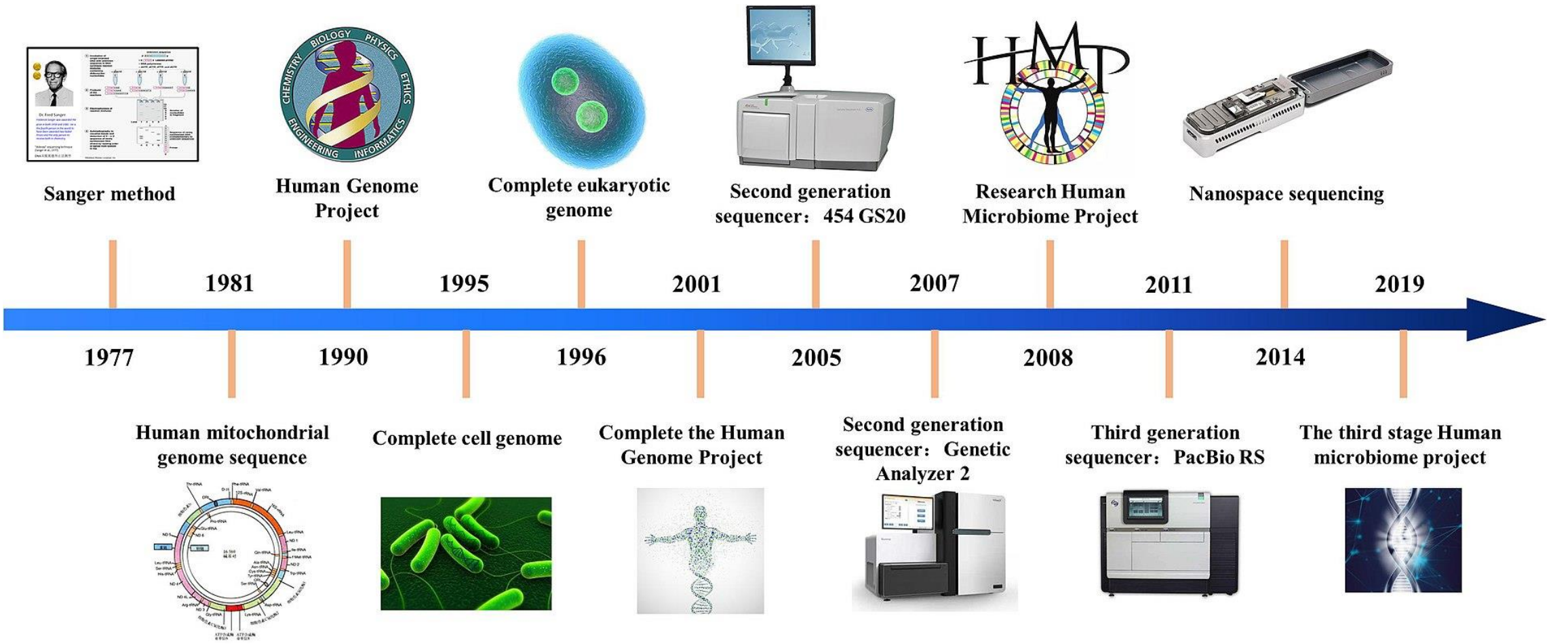
Cancer Genome Somatic Variation



Mutation burden in 20 tumor types and relative contribution of different mutational processes.
For each tumor type, samples were divided into deciles on the basis of their mutation burden. (2015)

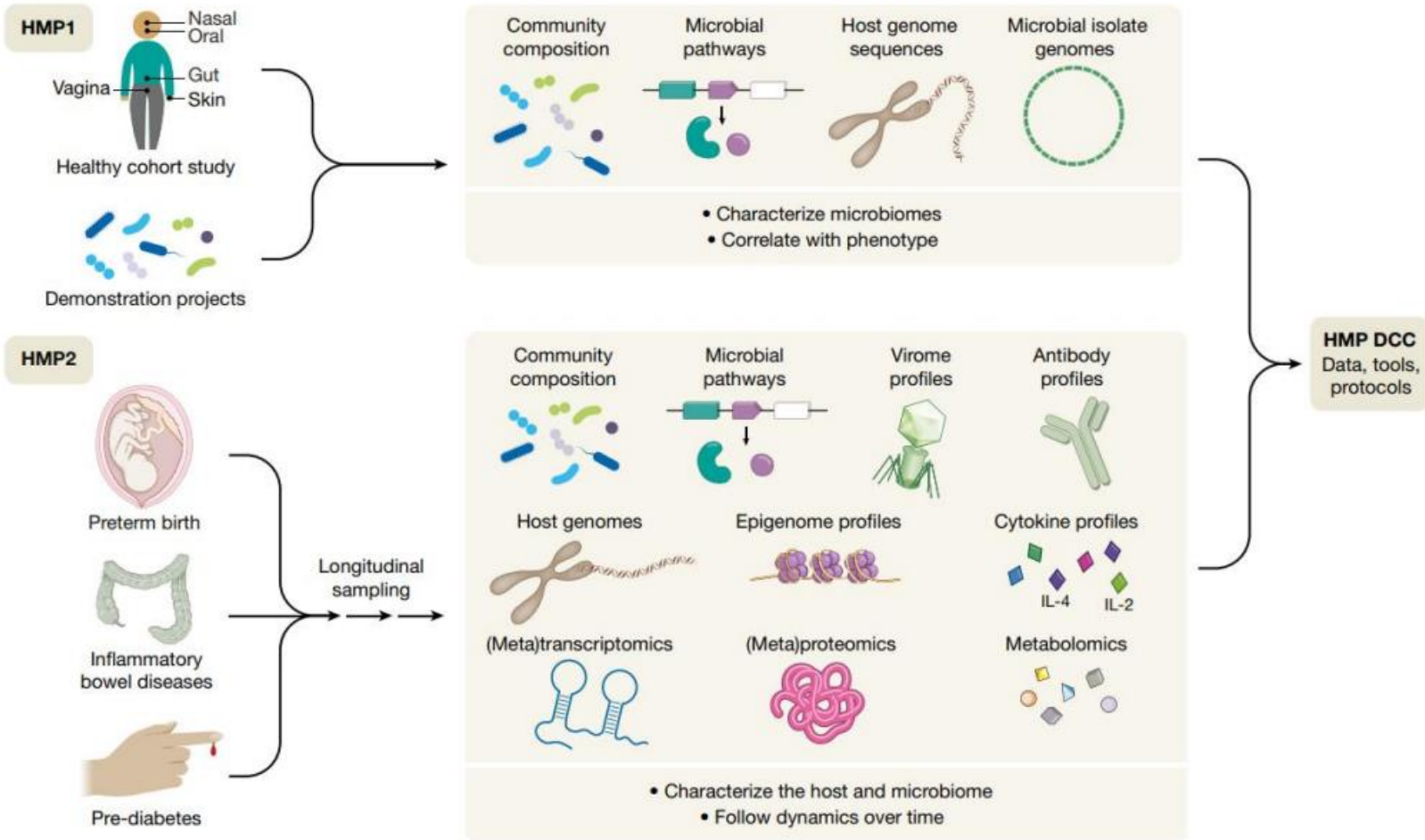
DOI: 10.1126/science.aab4082

High-throughput sequencing (HTS) methods



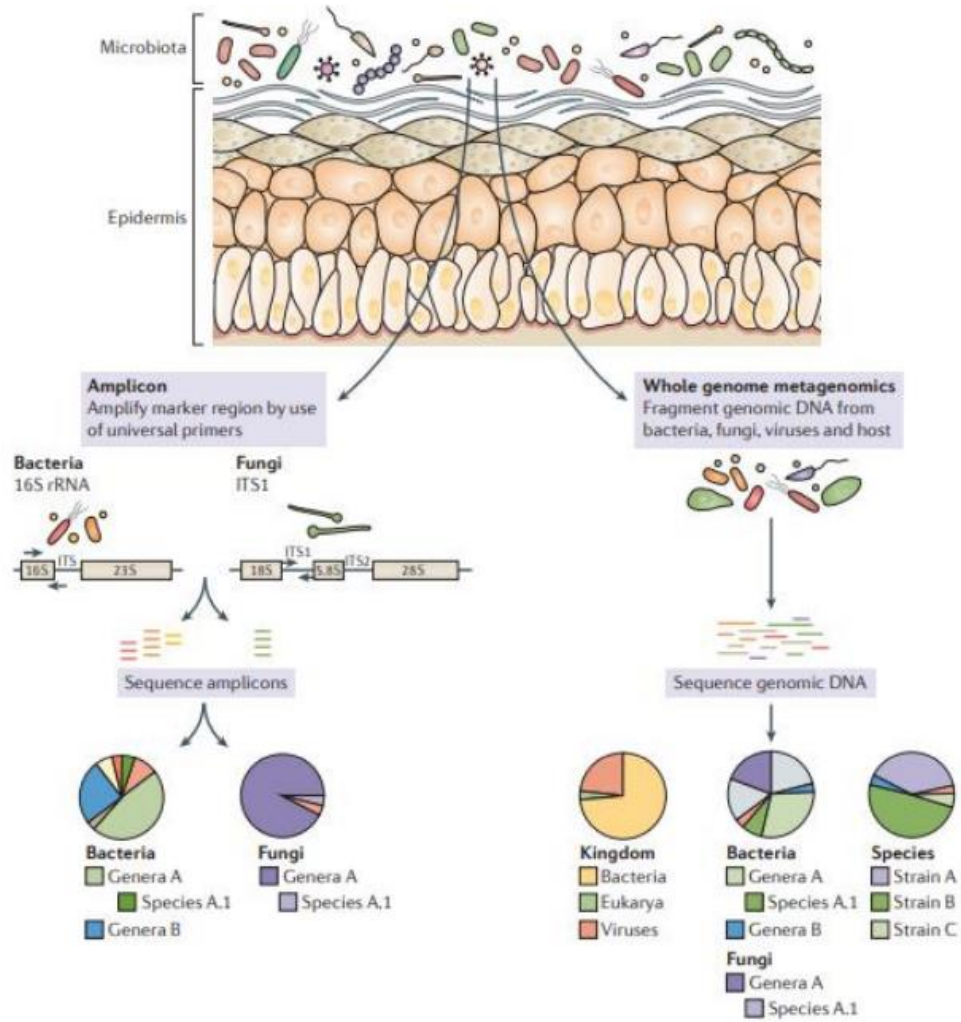
https://en.wikipedia.org/wiki/DNA_sequencing

The first and second phases of the NIH Human Microbiome Project

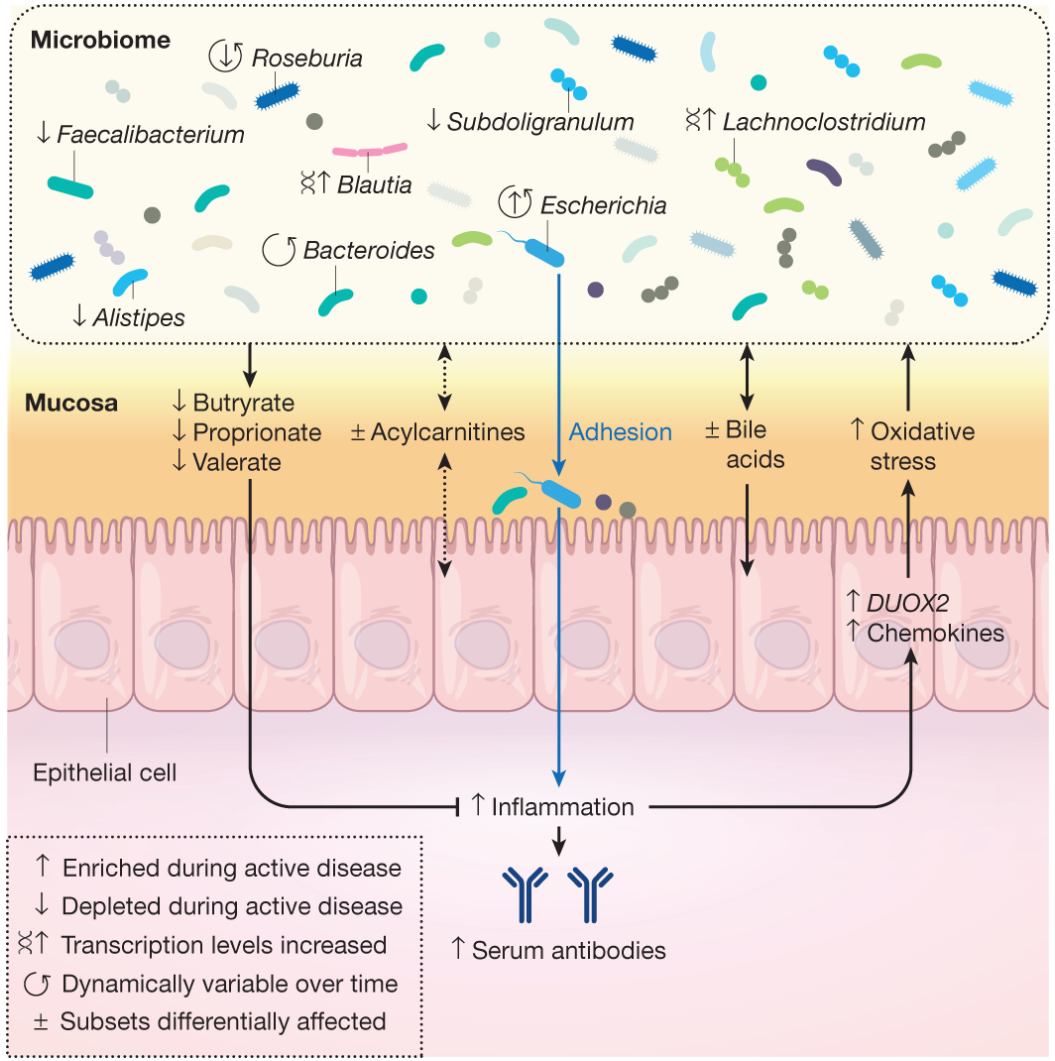


https://www.nature.com/articles/s41586-019-1238-8

Human Skin Microbiome Project

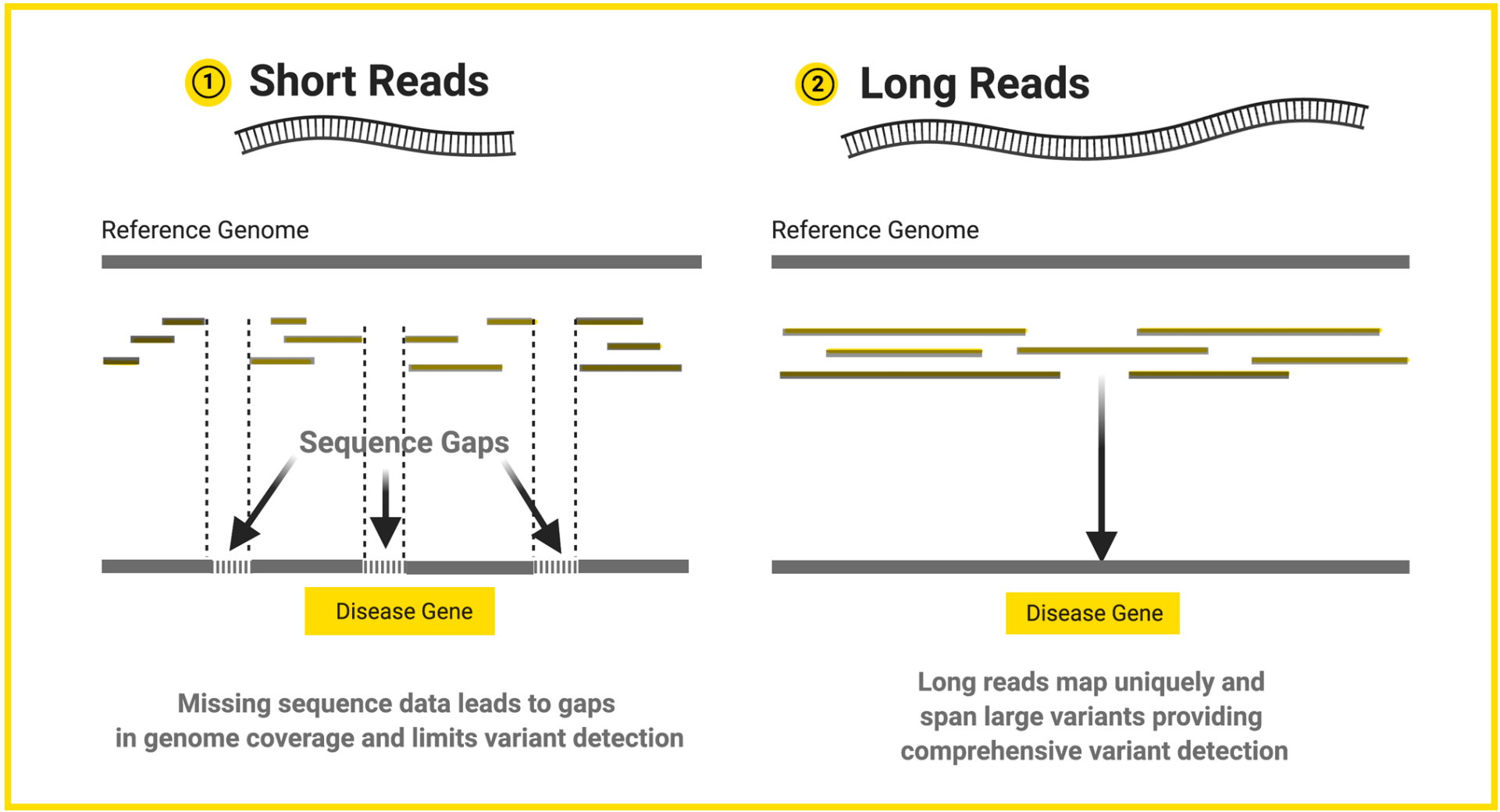


Host–microbiome dynamics in inflammatory bowel diseases



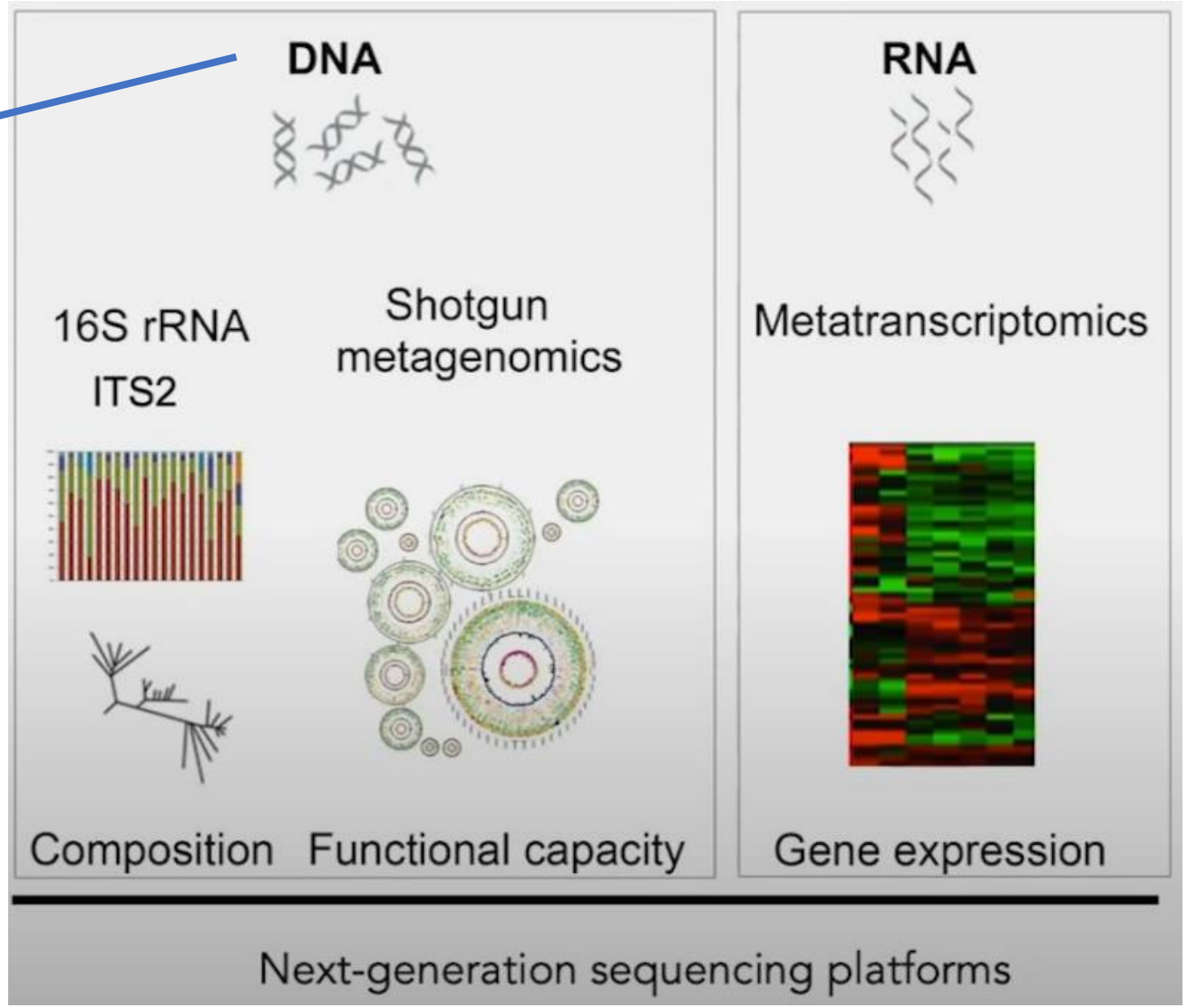
https://www.nature.com/articles/s41586-019-1238-8

Short read vs Long read

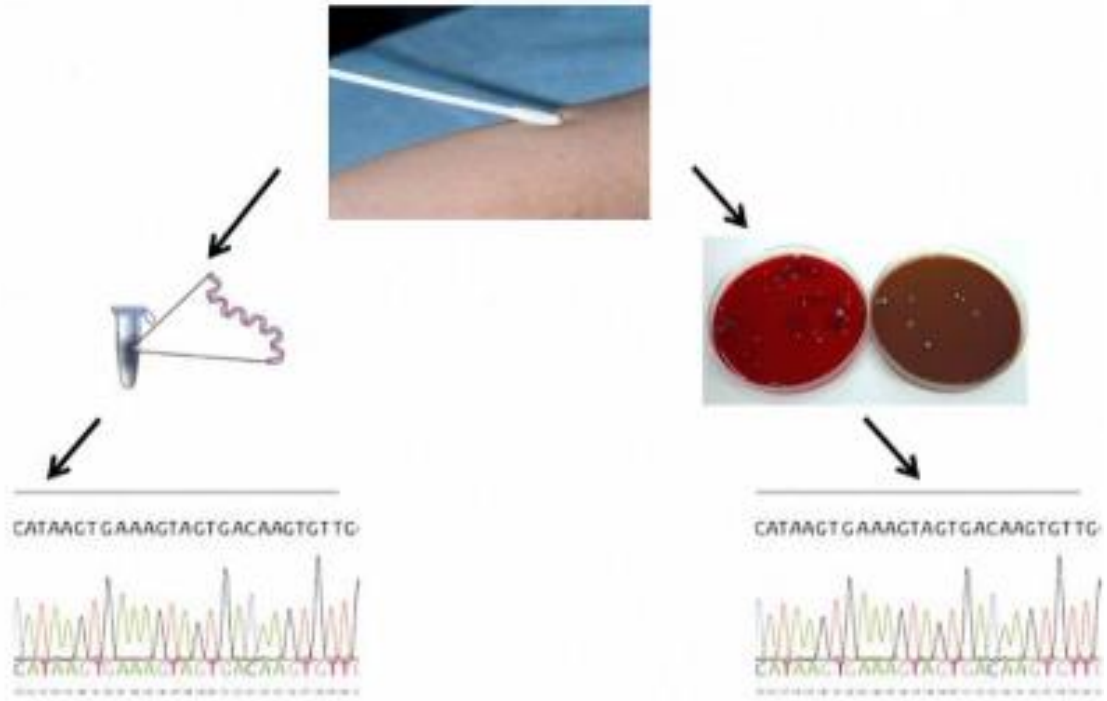


NGS Applications in microorganism research

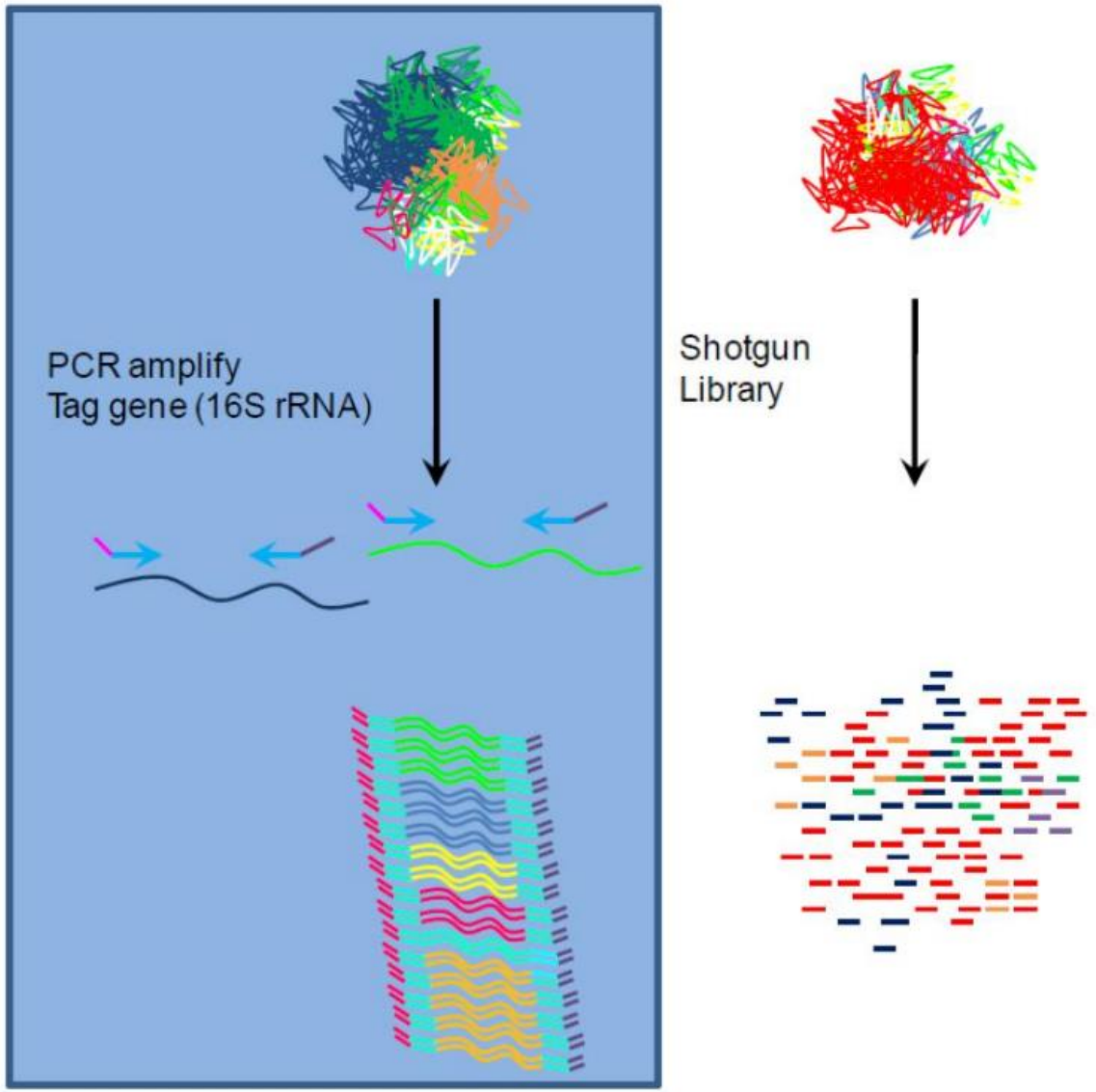
Whole Genome Sequencing (WGS)



Direct sequencing vs. culture-based methods



Target Gene vs Metagenomics



NGS Applications in microorganism research

- **DNA-seq**:
  1. Target sequencing (some genes): serotyping and genotyping
  2. Whole Genome Sequencing (WGS): multi locus genotyping, genetic diversity and evolution study
  3. Amplicon metagenomics (16S/ITS): identification/classification/ surveillance study
  4. Shotgun metagenomics: resistome
- **RNA-seq**:
  1. Transcriptomics: gene expression for a isolate
  2. Meta-transcriptomics: gene expression for a bulk sample
  3. Single cell RNA-seq: gene expression for individual cells

Thank you for your attention!