## Introduction to Genome Sequencing

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#### How do we sequence bacterial genomes?



Technology	Machine	Read length	Output from one run	Error Rate
Illumina	HiSeq	75-150bp	250-300Gb	<1%
	MiSeq	75-300bp	15Gb	
Ion Torrent		200-400bp	2Gb	~1%
PacBio		Tens of kb	Up to 10Gb	~15%
Oxford Nanopore		Tens of kb, up to 1Mb	~15Gb	~15%

## **Illumina and Ion Torrent**

- Cut DNA into short fragments
- Amplify
- Add bases and detect incorporation

#### Illumina sequencing - amplification



Images from www.illumina.com

#### Illumina sequencing – detection



#### **Illumina sequencing - detection**



Images from illumina.com



https://www.flickr.com/photos/97397973@N05/19092162353/

## Ion Torrent Sequencing – amplification

One DNA molecule per bead. Clonal amplification to thousands of copies occurs in microreactors in an emulsion



#### Ion Torrent Sequencing – detection



Image from Mardis, E.R. Next-Generation Sequencing Platforms. Annu. Rev. Anal. Chem. 2013. 6:287–303



https://www.flickr.com/photos/fdaphotos/8754348757/

# PacBio and Nanopore - single molecule sequencing

- No amplification needed
- Reduces GC bias
- Allows for longer reads
- Currently higher error rates

## Pacific Biosciences (PacBio)



Images from Metzker, M.L. (2010). Sequencing technologies - the next generation Nat. Rev. Genet. 11, 31-46.



https://www.flickr.com/photos/doe\_jgi/3481004 5553/

#### **Nanopore Sequencing**



## **Nanopore Sequencing - detection**

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Loman, Nicholas (2014): Wiggle plot showing Oxford Nanopore signal data for a P. aeruginosa read. figshare.

