

# Ecological Genomics Notes

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## January 23, 2017:

### Info Update:

- Advantages in Seq tech.
- Range of applications:
  - WGS (whole genome sequencing)
  - RNAseq (sequencing RNA converted to cDNA)
  - Targeted capture seq. (string of probes mixed with sample, pulls immune related genes from organism and seq just those)
  - ChIPseq (chromatin immunoprecipitation seq, recognizes and antibody and pulls out all DNA bound to that protein)

### Why one or the other?

Genetic variation

- phenotypes

number of samples

- population
- individual
- comparative studies
- model or not

Demographic history

Adaptive genetic variation

gene expression var.

length of reads

number of reads

distribution

Reads:

- short = 50bp
- long 100 bp, 150 bp, 300 bp (miseq)
- 10,000-60,000bp = SMRT

Single vs. paired end

- General library Prep. Workflow
  - extraction (DNA, RNA -> to cDNA)
  - fragment sample
  - ligate adaptors (individual barcodes)
  - add seq. adaptors

### *Reduced Rep*

- RNA -> coding
- GBS/RAD-seq
- near restriction sites
- Sequencing-by-synthesis (SBS)
  - bridge amp
  - cluster gen.
  - labeled dNTP (ATCG)
- Other Technologies
- Learning Activity

### **Human Genome Project (2001-2003)**

- ABI = Sanger
- 15 years
- 1 genome (one person)
- \$3 billion

Uses PCR and sequences broken by faulty base pairs to work backwards

### **2014 X-Ten releases**

- HiSeq by Illumina (look up video of how it works)
- 1 day
- 45 whole genomes
- \$1000 bucks each

Sheet of glass with 8 lanes with flow cells... (look up)

### **Take home messages**

- Likely using Illumina seq. (just library tech. changes) usually SBS
- adaptors are markers (barcodes) used to identify samples during sequencing
  - first thing that's sample and gives ID barcode = seq adaptor
  - alago is a sequence of DNA that is attached to plate binds to sample
- Model vs. non Model:
  - short reads (assembly to create long sequence based on short reads that shift)
  - denovo assembly -> computer program (added variability with mixed sample)
  - 15% error for SMRT can be reduced to less than 1 with repeated passes
  - illumina is much smaller but more accurate (0.05% error)
  - combine the two to have a higher degree of confidence

### **Paper Discussion:**

#### **Three advances in biology:**

- Modern Synthesis (evolution (Darwin) and population genetics (Mendel))
- Watson and Crick (molecular biology DNA)
- Omics Era (genomics, proteomics etc.)

## What do we think about this?

- Phylogenies -> reducing error bars or reshaping question?
- Do you throw out experimental design and scientific method for large scale shotgun blast sequencing?  
- > can still be used to do hypothesis driven science, but also something to be said for sending a telescope into deep space.
- Most journals and funding sources require all seq data be made public - leads to a storage space issue  
-> genbank ran out of space and needed emergency funding from congress.
- data are not reviewed very well some gen bank sequences are kinda rough
- reference genomes are a sample size of 1 so it might not be representative

On Wednesday two papers to talk about:

- 1) where do genomics data have limitations and why we use them
- 2) discussion leaders (sign up via blackboard)
- 3) next week talk about some of library preps (four of them) four different update people

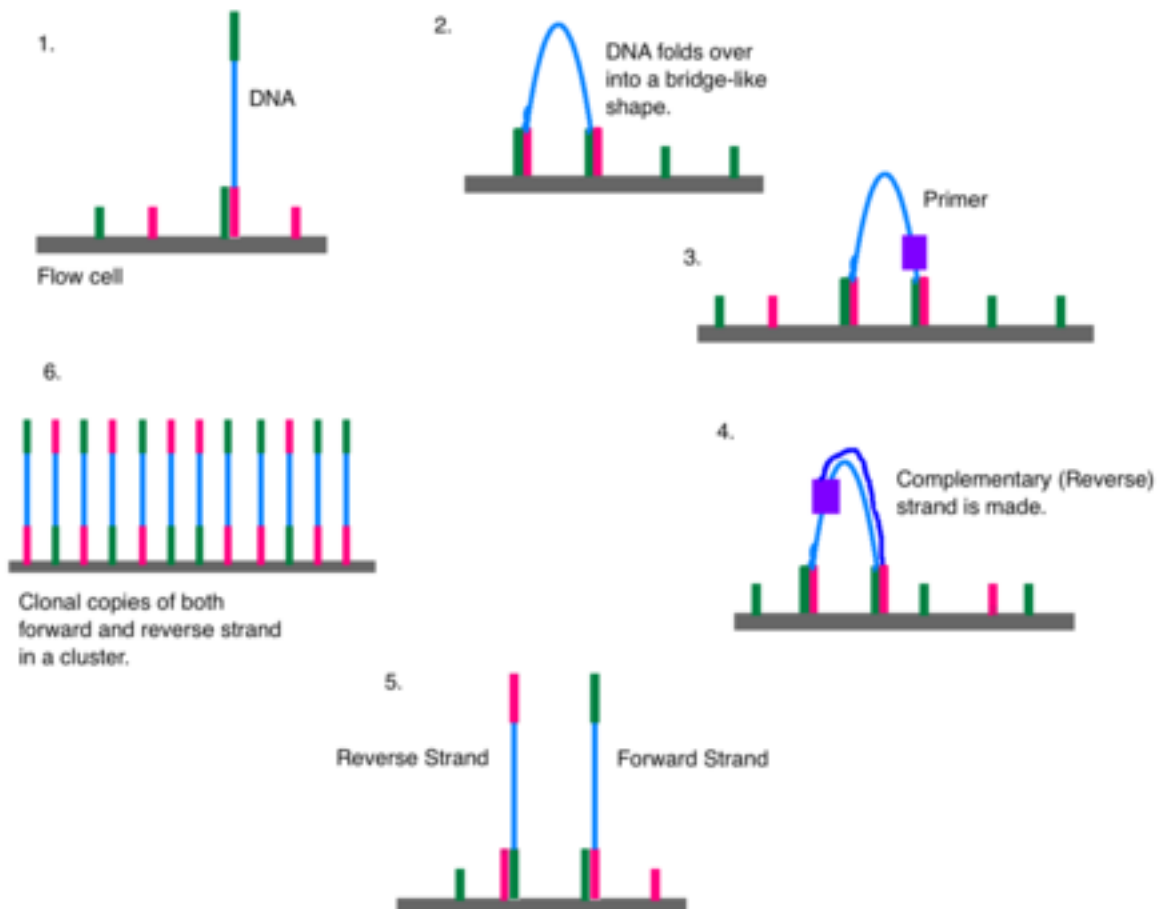


Figure 1: Illumina Sequencing