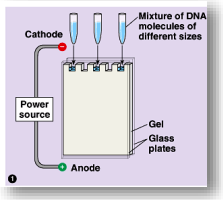
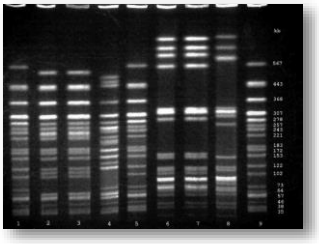
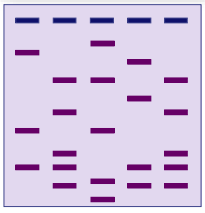


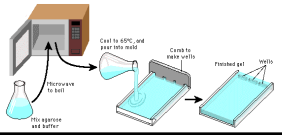
Biotechnology
Gel Electrophoresis
Part I: Basics

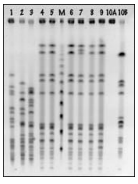




Comparing cut up DNA

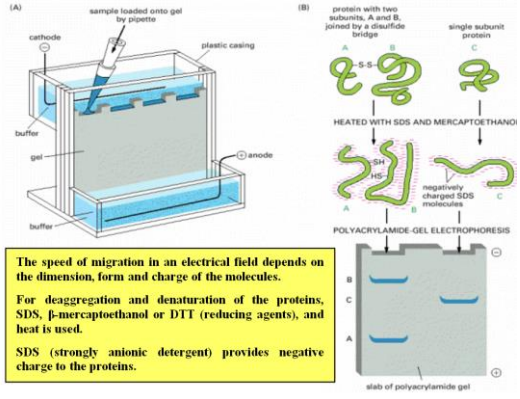
- How do we compare molecules?
  - separate fragments by size and/or charge
- How do we analyze DNA?
  - separate fragments by size
- How do we separate DNA fragments?
  - run it through a gelatin
  - gel electrophoresis





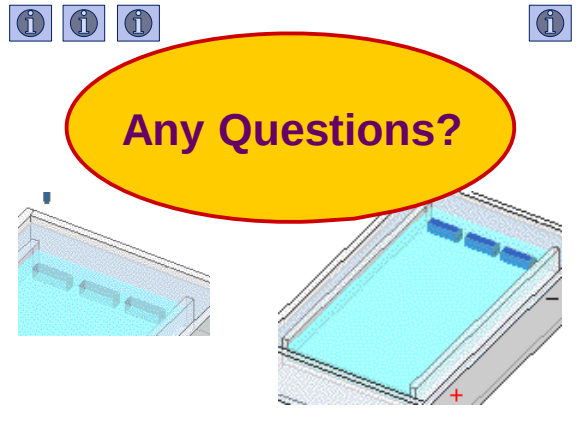
Comparing cut up DNA

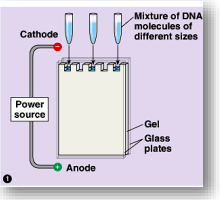
- How do we compare molecules?
  - separate fragments by size and/or charge
- How do we analyze proteins?
  - separate fragments by size and/or charge
- How do we separate proteins?
  - in agarose:
    - determine isoelectric point (native)
  - in polyacrilimide
    - SDS – PAGE (non-native)



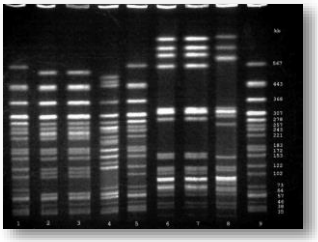
Components of Electrophoresis

- voltage
  - "pulling" power!
- buffers: TAE vs. TBE vs. other…
  - tris-acetate-EDTA
    - lower buffering capacity than TBE
    - slightly faster migration
  - tris-borate-EDTA
    - higer buffering capacity than TAE
    - slightly slower migration
- agarose vs. polyacrylimide
- time!



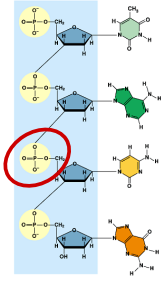


Biotechnology
Gel Electrophoresis
Part II: DNA



Gel electrophoresis

- A method of separating molecules (i.e. DNA) in a gelatin-like material using an electrical field
  - DNA is negatively charged
  - when it's in an electrical field it moves toward the positive side

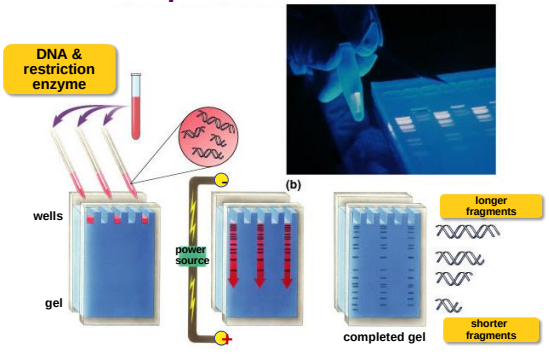


Gel electrophoresis

- charged particles move in an electrical field
  - with DNA fragments, size of affects how far it travels
  - [agarose] affects speed
    - small pieces travel farther/faster
    - large pieces travel less/slower

% Agarose concentration in gel (w/v)	Efficient separation range for linear double stranded DNA molecules (Kb)
0.3	5 - 60
0.6	1 - 20
0.7	0.8 - 10
0.9	0.5 - 7
1.2	0.4 - 6
1.5	0.2 - 3
2.0	0.1 - 2

Gel electrophoresis

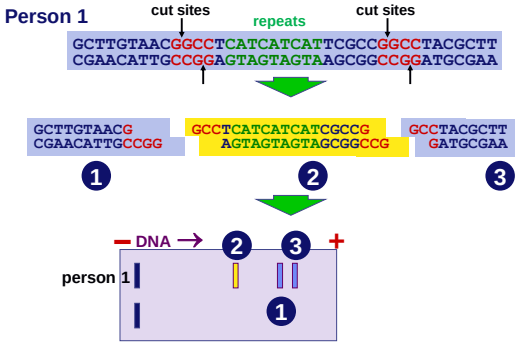


Uses: DNA fingerprint

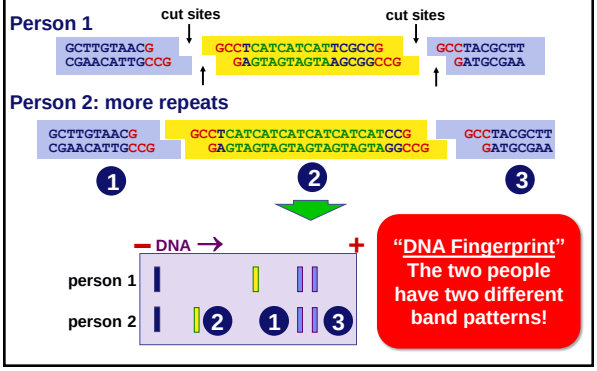
- Why is each person's DNA pattern different?
  - sections of "junk" DNA
    - doesn't code for proteins
    - made up of repeated patterns
      - CAT, GCC, and others
      - each person may have different number of repeats
    - many sites on our 23 chromosomes with different repeat patterns

Person 1	GCTTGTAACGGCCTCATCATCATTCGCCGGCCTACGCTT CGAACATTGCCGGAGTAGTAGTAAGCGGCCGGATGCGAA
Person 2	GCTTGTAACGGCATCATCATCATCATCATCCGGCCTACGCTT CGAACATTGCCGTAGTAGTAGTAGTAGTAGGCCGGATGCGAA

DNA patterns for DNA fingerprints

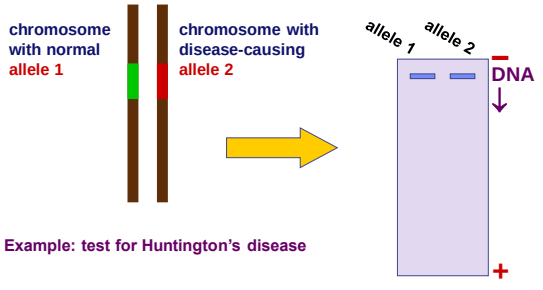


Differences between people



Uses: Medical diagnostic

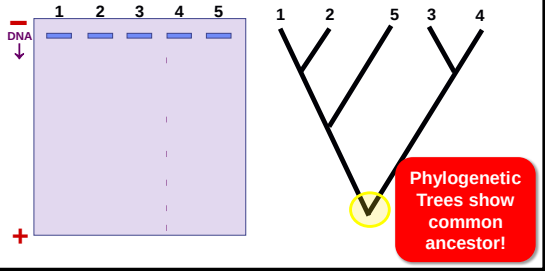
- Comparing normal allele to disease allele



Example: test for Huntington's disease

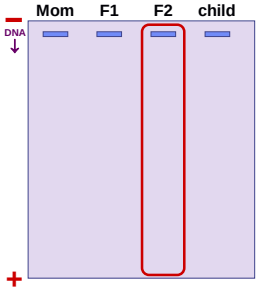
Uses: Evolutionary relationships

- Comparing DNA samples from different organisms to measure evolutionary relationships



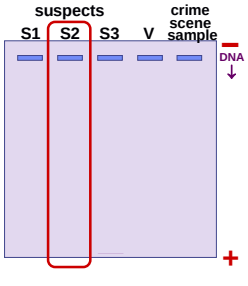
Uses: Paternity Testing

- Who's the father?



Uses: Forensics

- Comparing DNA sample from crime scene with suspects & victim



Uses: Forensics

- Comparing blood samples on defendant's clothing to determine if it belongs to victim
  - DNA fingerprinting