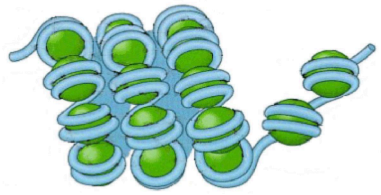
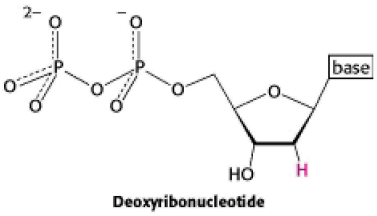
Crack the DAT

Crack Notes [Biology 2]
Genes

Genes

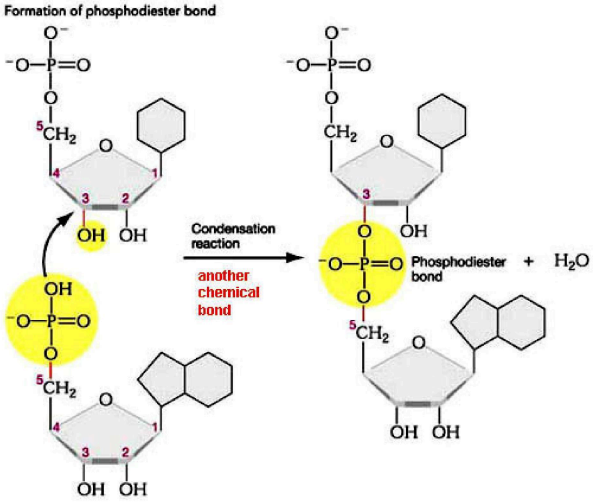


- Definitions
  - *Gene*: DNA that codes for a single polypeptide/mRNA/rRNA/tRNA
  - *Euchromatin*: region of DNA containing genes being actively transcribed
  - *Heterochromatin*: region of DNA containing genes not actively transcribed
  - *Genome*: entire DNA sequence of an organism — human: 26k-38k genes, ~1% codes for protein
  - *Central Dogma*: DNA —(transcription)—> RNA —(translation)—> protein



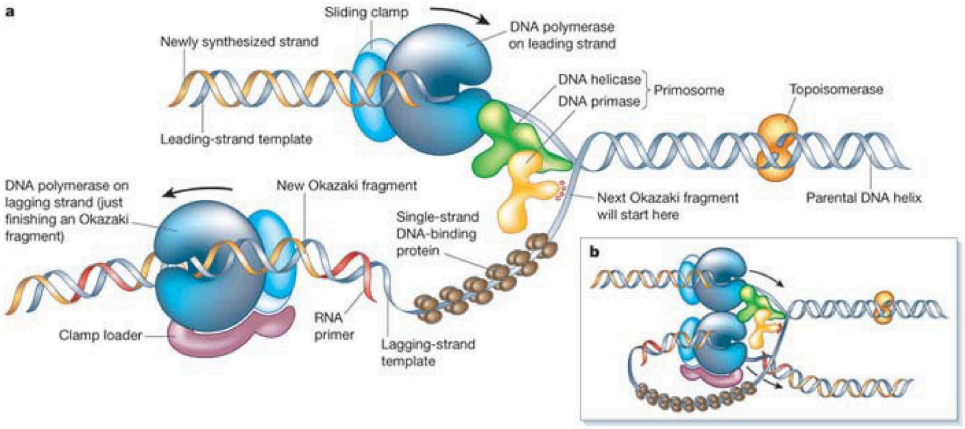
Deoxyribonucleotide

- DNA
  - *Nucleotides*: phosphate group + 5-carbon sugar + nitrogenous base
  - *Bases*: adenine (A), guanine (G), cytosine (C), thymine (T)
  - *Purines*: adenine & guanine, two rings
  - *Pyrimidines*: cytosine & thymine, one ring
  - Bases bound together by 5'-3' phosphodiester bonds into phosphate backbone
  - 5'→3' directionality (5' phosphate, 3' OH)
  - DNA strands are antiparallel, base pairing occurs between A=T, G≡C - "A2T C3G"
  - Double helix, complementary strands



Replication (DNA → DNA)

- Semiconservative — new double stranded DNA = 1 old strand + 1 new strand
- Replication begins at the origin of replication, proceeds in two directions (bidirectional)
- Continuous reading/replication can occur at leading strand, but not at lagging strand
  - Lagging strand is continuously interrupted & restarted (Okazaki fragments)
  - Typically fragments are in range of 10^2 - 10^3 bp



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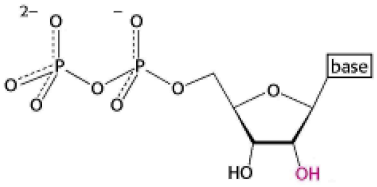
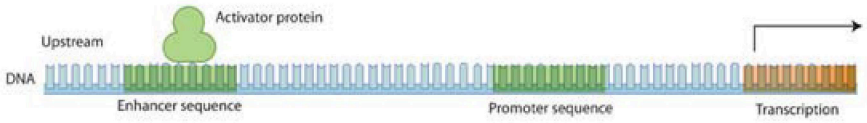
- Enzymes
  - *Helicase*: unzips the double helix, allows primase to bind
  - *DNA polymerase*: builds new DNA strand, can only add nucleotides to existing strand
    - Reads parent strand in 3'→5' (upstream), creates new strand in 5'→3' (downstream)
    - Also contains a mechanism to repair mismatched nucleotides, adds to accuracy
  - *Primase*: creates RNA primer so DNA polymerase can start working
  - *DNA ligase*: connects chunks of new DNA created in lagging strand
  - *Telomerase*: lengthens telomeres (repeated sequences at ends of eukaryotic DNA, protects from being eroded)

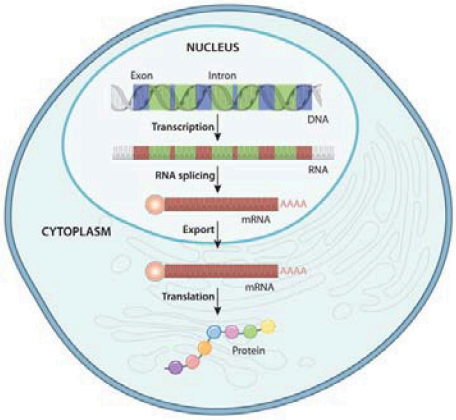
- RNA
  - 2' carbon on sugar has an OH instead of an H
  - Generally single stranded, though can have special secondary structures
  - Has uracil (U) instead of T, so A=U and C≡G bonds



Ribonucleotide

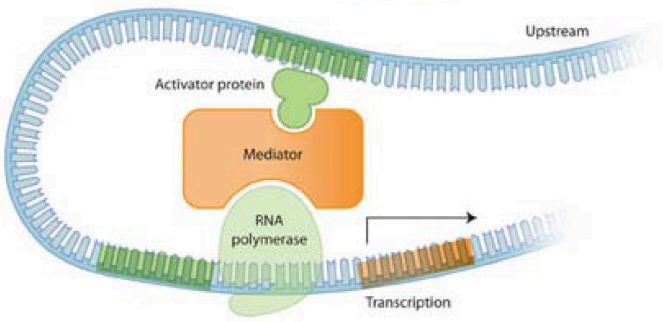
- Types of RNA
  - *mRNA* (messenger RNA): carries genetic info from nucleus (DNA) to cytosol
  - *rRNA* (ribosomal RNA): forms ribosomes by combining with proteins, made in nucleolus
  - *tRNA* (transfer RNA): carries amino acids to ribosomes





Transcription (DNA->RNA)

- *Initiation*: initiation factors recognize promoter, cause assembly of RNA polymerase
  - Can be regulated by small changes in promoter sequence, which affect binding
  - Can be regulated by activators and repressors which bind DNA close to promoter
  - RNA polymerase unzips double helix, transcribes only antisense (-) strand
- *Elongation*: no proofreading, slower than replication
  - RNA polymerase reads DNA in 3'→5' direction, creates RNA in 5'→3' direction
- *Termination*: RNA pol recognizes termination sequence



- Prokaryotes - gene activity changes as a response to changes in environment
- Eukaryotes - gene activity changes to maintain homeostasis

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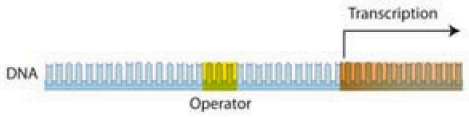
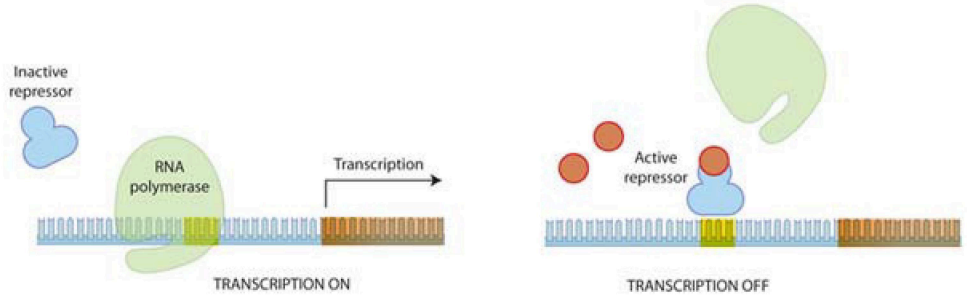
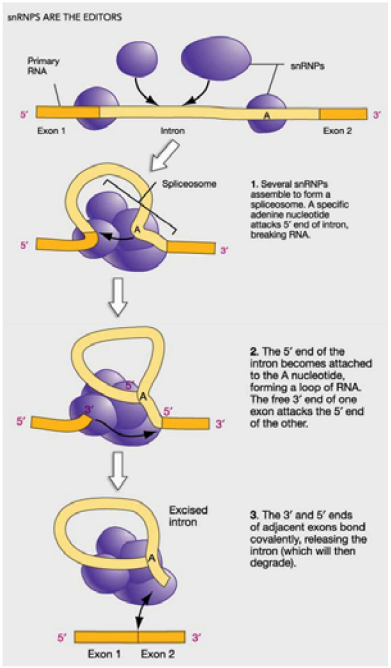


- Operon
  - Prokaryotic structure with operator, promoter, and multiple genes
  - *Operator*: binds repressor protein to prevent RNA polymerase from binding, or binds activator protein to facilitate RNA polymerase binding
  - *Promoter*: binds RNA polymerase in the absence of repressor
  - Multiple genes will appear on single transcript
  - *Lac operon*: repressor protein allosterically deactivated by lactose, so increase in lactose concentration leads to repressor not binding → transcription of lactose proteins



- Post-Transcriptional Processing (Eukaryotes)
  - Replication → primary transcript → processing
  - *5′ cap*: helps ribosome to attach, protects transcript from degradation
  - *poly-A tail*: on 3′ end, protects transcript from degradation
  - *Intron splicing*: snRNPs (small nuclear ribonucleoproteins) cleave out introns, leaving behind exons which are spliced together to form final transcript
  - Variation in this process can make different proteins for same transcript



Translation

- 4 different bases, 3 base sequence codes for each amino acid → 43 = 64 possibilities
- Since there's only 20 amino acids, more than one sequence can code for the same amino acid → genetic code is *degenerate*
- But each sequence can code for only one amino acid → genetic code is *unambiguous*
- 3 base sequence = *codon*
- UAA/UGA/UAG are *stop codons*, AUG is *start codon* but also codes for Met
- Ribosome has a small subunit and a large subunit, produced in nucleolus
- Each tRNA has an anticodon to specifically recognize codons

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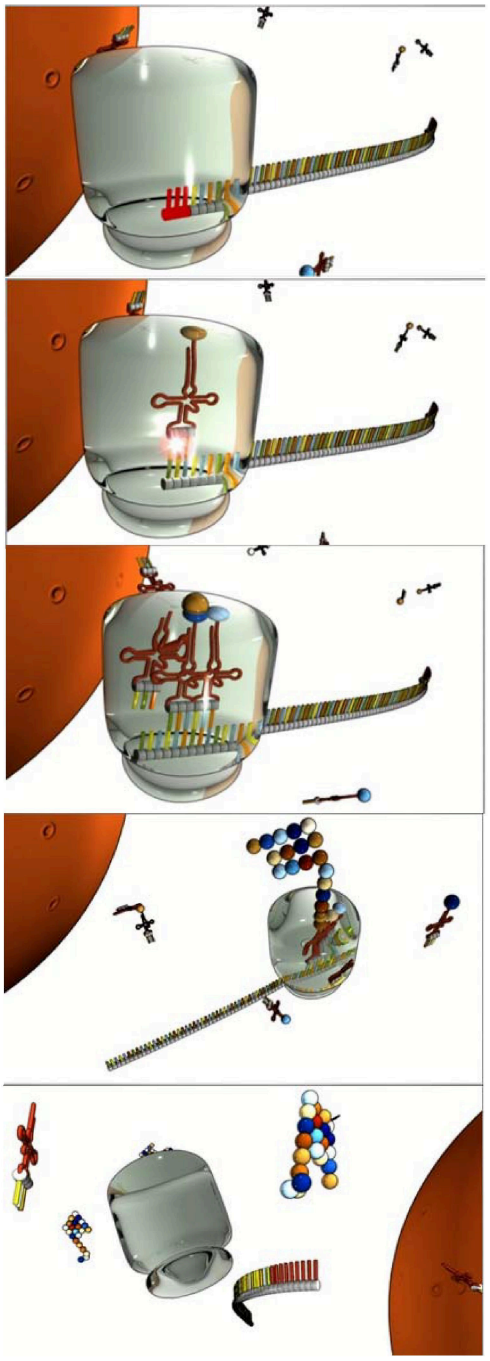
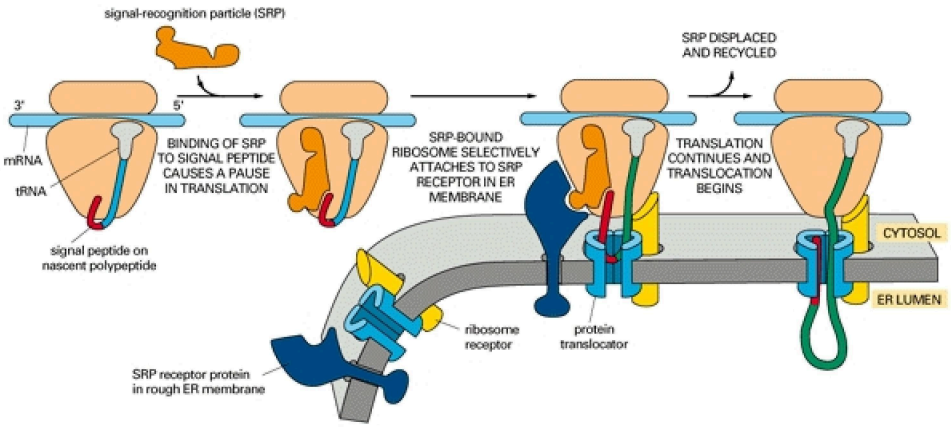
- Initiation
  a. mRNA recognized by small subunit of ribosome, binds together
  b. tRNA with 5'-CAU-3' anticodon (bound to Met) binds to start codon at P site of ribosome
  c. Large subunit binds

- Elongation
  a. tRNA binds to A site of ribosome
  b. Peptide bound to tRNA at P site gets attached to amino acid at A site
  c. Translocation occurs; tRNA at P site shifted to E (exit) site and leaves, tRNA at A site now attached to peptide moves to P site

- Termination
  a. Stop codon reaches A site
  b. Release factors recognize it and cause disassembly of ribosome

- Post-translational modifications can also occur where other stuff is attached to amino acids
  - If ribosome stays floating in cytosol → protein ends up in cytosol
  - If peptide contains a special signal peptide, a SRP will carry ribosome & peptide to a receptor on the endoplasmic reticulum, causing the ribosome to attach → protein ends up inside ER, will become membrane bound proteins or exported from the cell





Mutations

- *Gene mutation*: change in DNA sequence within a gene
- *Chromosomal mutation*: change in chrosome structure
- *Mutagen*: something that increases the chance of mutation
- *Forward mutation*: mutated organism mutating again
- *Backward mutation*: mutation that cancels another mutation
- *Wild type*: non mutated organism

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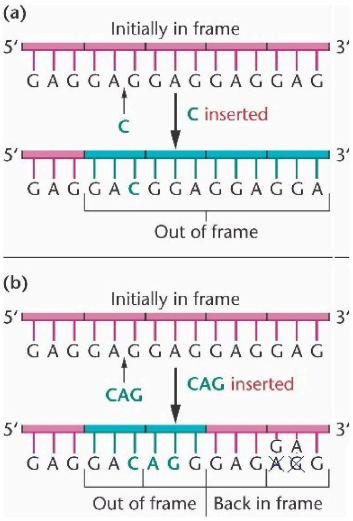
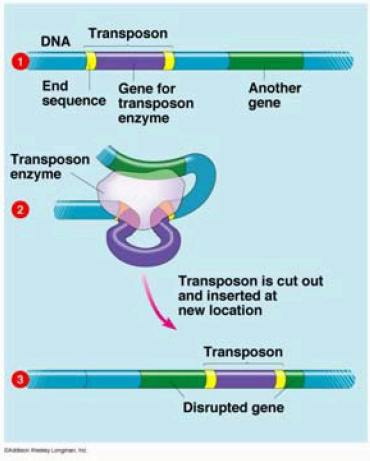
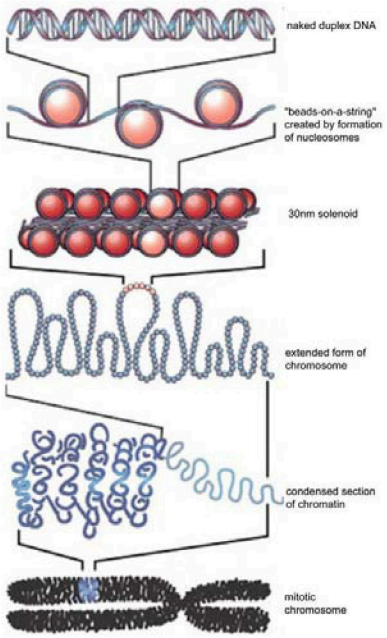
- Gene Mutations
  - *Point mutation*: one base pair changes, i.e. base-pair substitution, insertion/deletion
  - *Missense mutation*: a base pair mutation that occurs in an exon, effects will vary
  - *Frameshift mutation*: if insertion/deletion occurs in multiples other than 3 → normally results in nonfunctional protein
  - *Nonsense mutation*: any mutation that creates a stop codon → nonfunctional protein





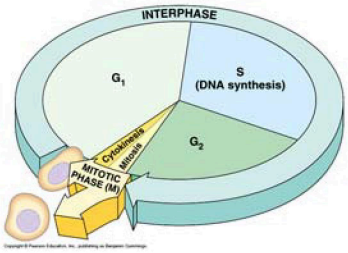
- Chromosomal Mutations
  - Deletions and duplications
  - *Translocation*: segment from one chromosome inserted into another
  - *Inversion*: orientation of DNA reversed on a chromosome
  - *Transposons* are DNA segments that are able to excise themselves from chromosome, reinsert themselves at another location

Cancer
- Unrestrained/uncontrolled growth of cells
- Normal cell growth genes can be converted into oncogenes by carcinogens



Chromosomes
- DNA wrapped up by *histone* proteins to form *nucleosomes*
- Nucleosomes then coil up into *chromatin*
- Humans have 46 separate dsDNA molecules, each called a *chromosome*
- Each chromosome has a partner that codes for the same traits called a *homologue*
  - Therefore humans have 23 homologous pairs of chromosomes
- *Diploid* (2n): a cell that has homologous pairs - all somatic cells have 23x2=46 chromosomes
- *Haploid* (n): a cell that doesn't have homologous pairs - all germ cells have 23 chromosomes

Cell Life Cycle
- Cell life cycle: "Go Sally, Go Make Children" or G1 -> S -> G2 -> Mitosis -> Cytokinesis
- G1: cell grows in size, can lead to G0 which is nondividing stage
- S: DNA replicates, chromosomes change from looking like > to looking like X
- G2: organelles duplicate, cell prepares to divide
- M: cell divides
- C: cytoplasm splits in two



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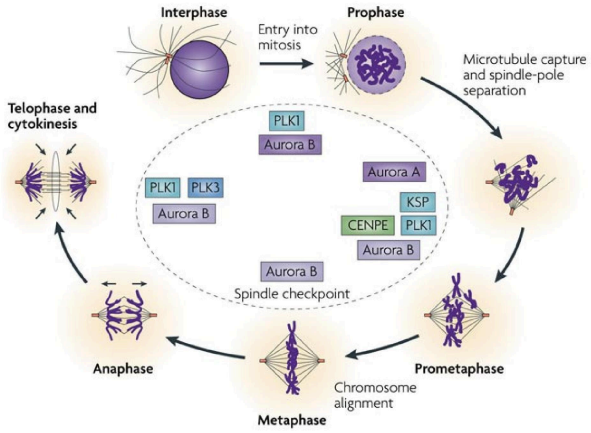


Mitosis - 1 diploid parent → 2 diploid daughter cells

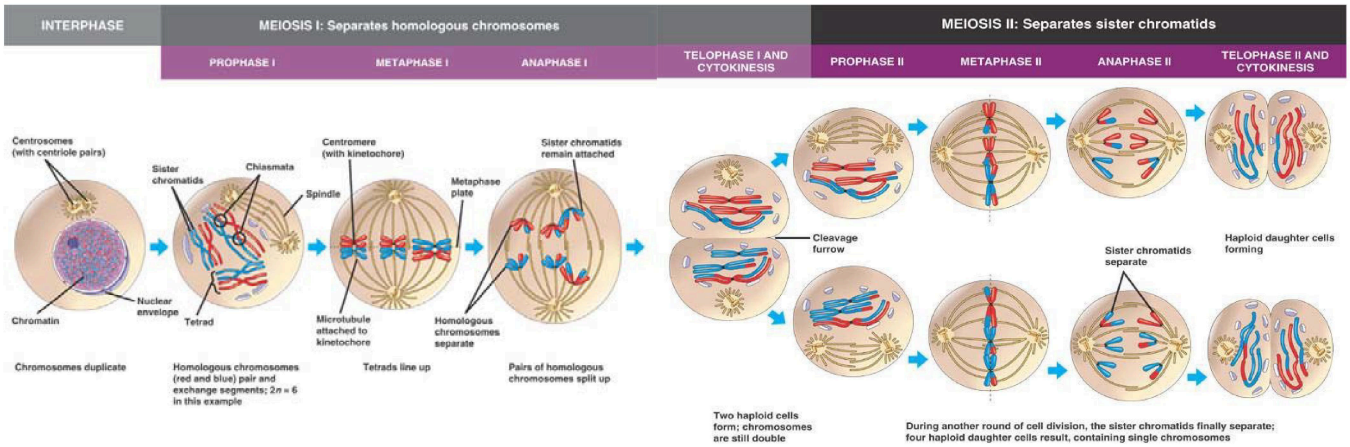
- *Prophase* - chromatin condenses into chromosomes, centrioles move to opposite sides, mitotic spindle begins to form, microtubules from each centriole attach to opposite sides of chromosomes at the kinetochore, nuclear membrane disappears, nucleoli disappear
- *Metaphase* - chromosomes align at middle of cell
- *Anaphase* - chromosomes split in half
- *Telophase* - chromatin decondenses, nuclear membrane reforms
- *Cytokinesis* - cytoplasm separates into two cells

Meiosis - 1 diploid parent → 4 haploid gametes/germ cells

- After S phase: primary spermatocyte/primary oocyte
- *Meiosis I* - primary spermatocyte turns into two haploid secondary spermatocytes, primary oocyte turns into one haploid secondary oocyte, polar body
- *Meiosis II* - 2 secondary spermatocytes turn into 4 spermatids. Only happens after fertilization for secondary oocyte, which turns into a diploid zygote + another polar body

- *Prophase I*: homologous chromosomes line up next to each other to form tetrads (XX), crossing over or genetic recombination occurs
- *Metaphase I*: XX lines up at equator
- *Anaphase I*: two X's split, travel towards opposite ends of cell
- *Telophase I*: nuclear membrane may reform
- *Cytokinesis*: primary spermatocyte splits evenly into secondary spermatocytes, but primary oocyte splits into a very small polar body and large secondary oocyte
- *Prophase II/Metaphase II/Anaphase II/Telophase II* occur similar to mitosis

- *Nondisjunction*: if centromere of any chromosome doesn't split in Anaphase I or II, results in cells having not enough/extra chromatids, common source of genetic diseases



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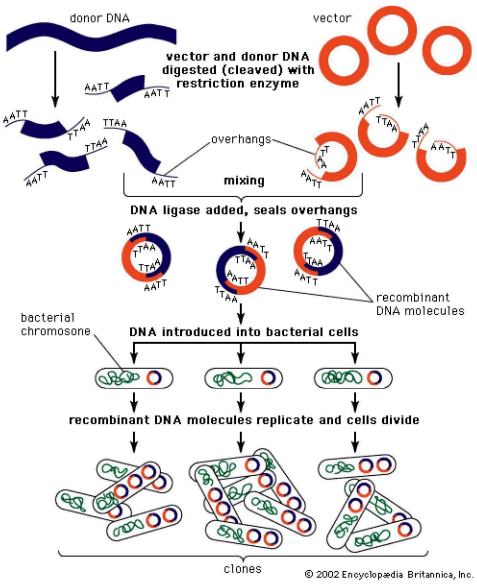
DNA Technology

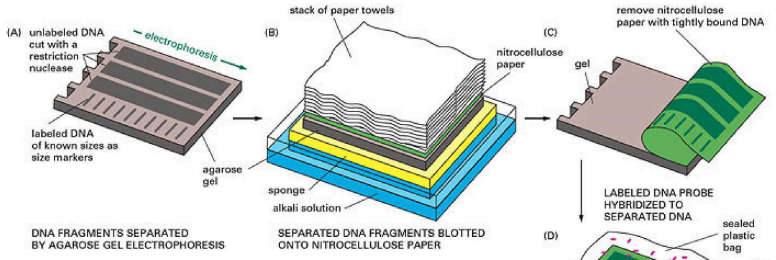
- Heating/adding salt can cause *denaturation*, higher % G-C needs high temp
- *Nucleic acid hybridization* used to identify unknown sequences by binding known sequences

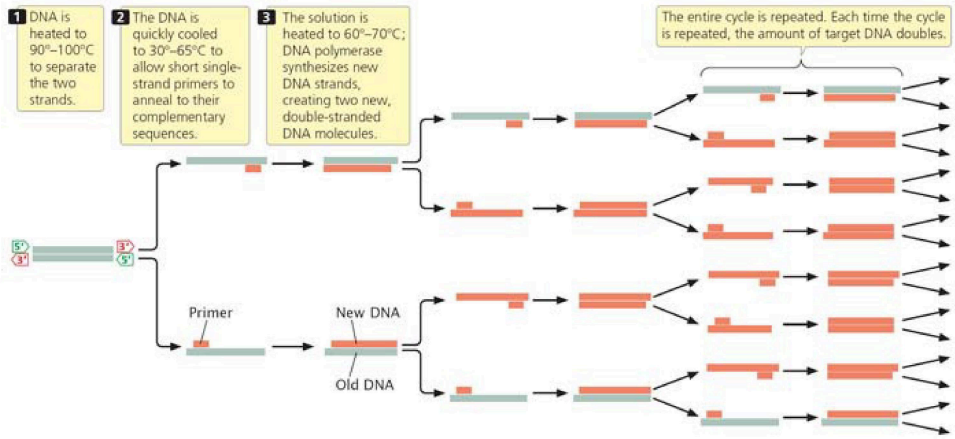


- Restriction enzymes
  - *Restriction enzymes* cut DNA at specific palindromic sequences
  - Can combine DNA cut with the same restriction enzymes to make *recombinant DNA*
  - Can add recombinant DNA to bacteria using a vector such as a plasmid or a virus, replicate a bunch of times to produce a *clone* containing the DNA, clones can be added to a clone *library*
  - If recombinant DNA contains an antibiotic resistance gene, the resulting clones can be screened by adding the antibiotic and keeping what's left
  - Can also search a library for a desired DNA sequence by adding a *probe*, a labeled complement to that sequence

- *Polymerase chain reaction*: can amplify specific double stranded DNA sequence using many copies of DNA primers. Process multiplies the # of desired dsDNA by two every time

- *Southern blot*: identifies specific sequences of DNA by chopping up DNA, separating pieces using gel electrophoresis, then adding probe





- *Northern blot*: same as Southern except for RNA
- *Western blot*: detects specific proteins using antibodies

- To replicate eukaryotic genes in bacteria, must start with eukaryotic mRNA which doesn't have introns, make *complementary DNA* (cDNA) with *reverse transcriptase* then add it to the bacteria

- *RFLP analysis*: identifies specific people based on distances between restriction sites