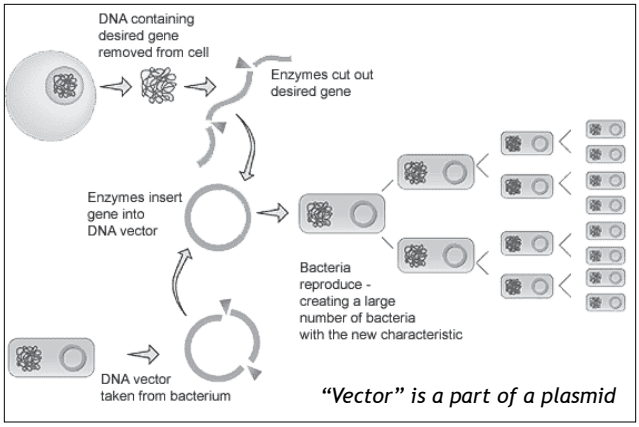
Biology AP
Cell Processes and Genetics

By Jennifer Zheng, David Guan. Edited by John Ho.

GENETIC ENGINEERING

Genetic engineering allows desired genes and proteins to be duplicated.

Process and Purpose Insert a foreign gene into a host cell that would reproduce gene of interest by transcription and translation using rna polymerase and ribosomes of host cell.	
Steps	1. Obtain gene of interest 2. Insert gene into host 3. Allow host to multiply gene
Plasmids	Circular DNA used in genetic engineering That gives bacteria additional traits (such as glow). It can be exchanged between bacteria



"Vector" is a part of a plasmid

- GREEN FLUORESCENT PROTEIN (GFP):
  1. GFP Gene: Codes for GFP protein.
  2. Bla Gene: Codes for enzyme ß-lactamase, which destroys antibiotic ampicillin. It serves as selectable gene.
  3. araC gene: Codes for araC protein.
- CONCEPTS:
  1. LB Broth is food for E. Coli.
  2. Ampicillin is an antibiotic and kills E. Coli unless plasmid (+DNA) is available.
  3. E. Coli with plasmid are transformed and are resistant to ampicillin

TRANSFORMATION

Process where foreign DNA (recombinant plasmid) is inserted into host cells. It is not a perfect process; only some of the final cells contain the plasmid.

- STEPS:
  1. Recombinant plasmids and host cells are mixed together
  2. CaCl2 is added. The ions neutralize negative charges on plasmid DNA and help plasmid to enter host cell.
  3. Heat shock rapidly changes temperature of solution, opening temporary pores in membrane which plasmids use to enter host cell
- WHY E. COLI: Reproduces quickly, nonpathogenic (harmless), has a fully characterized genome (all genes have been sequenced).
- SELECTION: Process that isolates the transformed E. Coli cells. Use selectable markers that show if plasmid is present inside E. Coli, such as penicillin, which kills all untransformed E. Coli.

RESTRICTION ENZYMES

Restriction enzymes cut DNA at specific nucleotide sequences. Used to find size of plasmid, number and location of restriction sites.

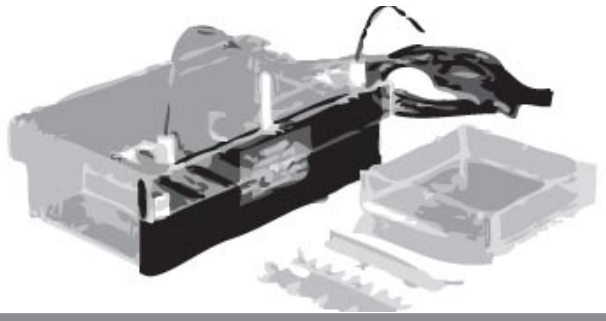
- BONDS: They break covalent bonds within single strands and hydrogen bonds between strands, as a result of coming apart.
- ENDS: Two different ends
  1. Blunt Ends: "chops" of DNA
  2. Sticky Ends: single stranded at ends, they are more beneficial because they can attach to other DNA fragments, resulting in recombinant DNA.
- STEPS: 1) Gene of interest and plasmid are cut with same restriction enzyme, 2) then mixed together. 3) DNA ligase (forms covalent bonds between nucleotides) is finally added to seal DNA back together.

GEL ELECTROPHORESIS

Gel electrophoresis technique used to separate and examine DNA fragments

1. DNA is cut with restriction enzymes.
2. DNA is added to wells at negative end of gel.
3. Electric current is turned on and migration begins.
4. DNA is separated on basis of size with help of electric charge (since larger pieces travel slower).

*Technique can be used to sequence DNA and determine order in which nucleotides appear



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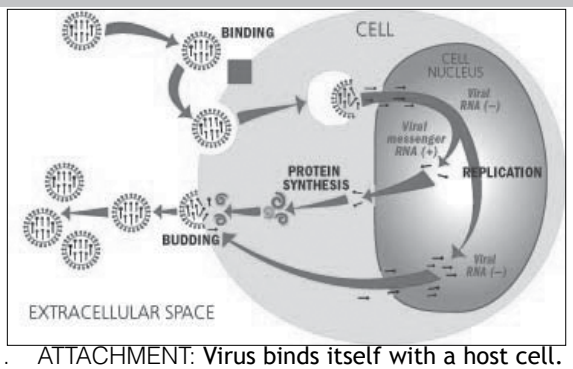
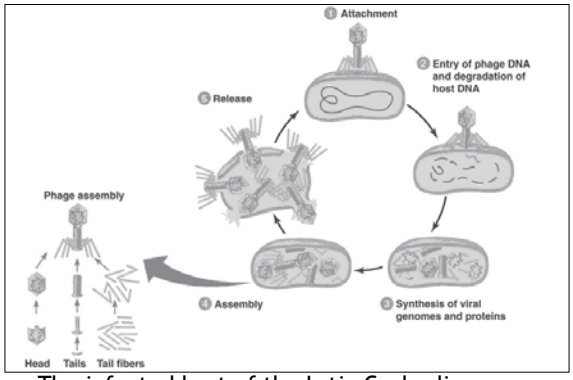
VIRUS REPRODUCTION



1. ATTACHMENT: Virus binds itself with a host cell.
2. ENTRY: The virus enters host cell.
3. BIOSYNTHESIS: Virus uses host cell to replicate own genes.
4. ASSEMBLY: Reproduced viruses assemble.
5. RELEASE: Assembled viruses leave cell.

- RETROVIRUSES: Also known as RNA viruses due to the fact they contain RNA instead of DNA. They also contain Reverse Transcriptase and creates DNA from its RNA, then uses the DNA to synthesize more RNA.

LYTIC CYCLE



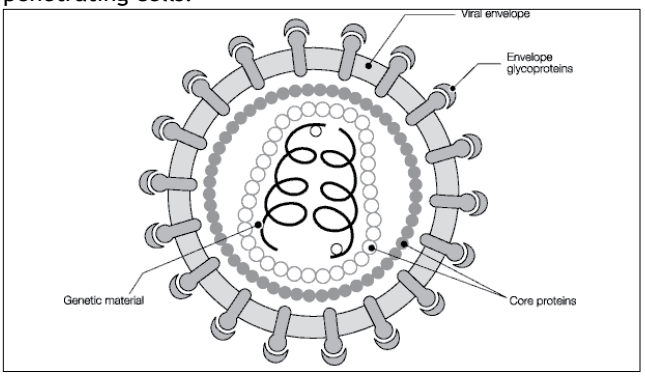
- The infected host of the Lytic Cycle dies.
- Bacteriophage who reproduce by only the lytic cycle are called “virulent phages”.

GENE THERAPY

EX VIVO	IN VIVO
• Takes a cell outside the body. • Adds a functional replacement gene into the cell. • Re-inserts the cell into the body. • Currently been proven to successfully treat hemophilia and A.D.D.	• Inserts genes with replacement genes directly into the body. • Currently in experimental stages, but may be used to cure cancer.

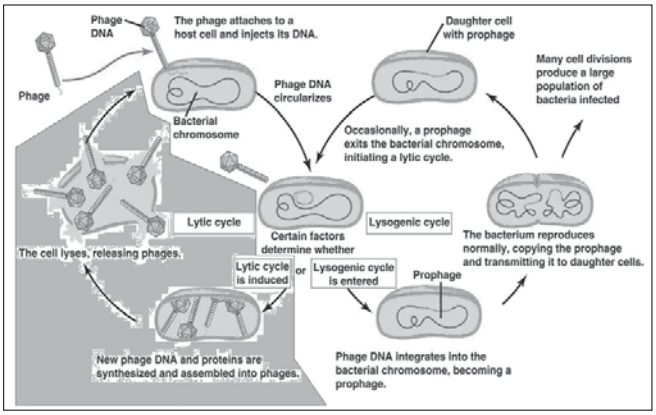
VIRUS STRUCTURE

- NUCLEIC ACID: Either composed of RNA or DNA.
- CAPSID: Also “protein coat”, it protects the nucleic acid.
- VIRAL EVELOPE: Serves to camouflage and assist the virus in penetrating cells.



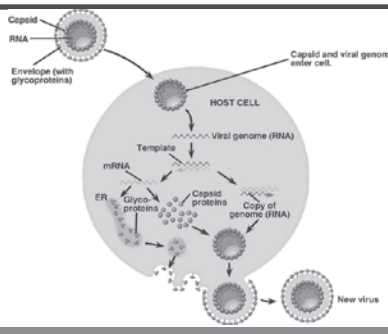
LYSOGENIC CYCLE

- During the Lysogenic Cycle the bacteria does not die
- Bacteriophage who can use either the lytic or lysogenic cycles are called “temperate phage”.



VIROIDS	PRIONS
• Viroids are circular RNA molecules that infect plant cells. • Like prions, they replicate by converting other proteins around them.	• An infectious protein that causes disease by making nearby proteins misfold. • They causes diseases in the brain and spinal cord.

ANIMAL VIRUSES



1. These viruses have Viral Envelopes to enable entry.
2. The glycoprotein attaches to a receptor on the host cell.
3. The virus enters the cell through endocytosis.
4. The viruses goes through Biosynthesis to “reproduce”.
5. New viruses assemble .
6. New viruses “bud” out of the cell, each taking a bit of the viral envelope.