

# CHAPTER 18 MICROBIAL MODELS: THE GENETICS OF VIRUSES AND BACTERIA

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## I. Introduction

- a) *Viruses and bacteria are the simplest biological systems - microbial models where scientists find life's fundamental molecular mechanisms in their most basic, accessible forms.*
- b) *Microbiologists provided most of the evidence that genes are made of DNA, and they worked out most of the major steps in DNA replication, transcription, and translation.*
- c) *Viruses and bacteria also have interesting, unique genetic features with implications for understanding diseases that they cause.*
- d) *Bacteria are prokaryotic organisms.*
- e) *Their cells are much smaller and more simply organized than those of eukaryotes, such as plants and animals.*
- f) *Viruses are smaller and simpler still, lacking the structure and most metabolic machinery in cells.*
- g) *Most viruses are little more than aggregates of nucleic acids and protein - genes in a protein coat.*

## II. The Genetics of Viruses

### A. Researchers discovered viruses by studying a plant disease

1. The story of how viruses were discovered begins in 1883 with research on the cause of tobacco mosaic disease by Adolf Mayer.
  - a) *This disease stunts the growth and mottles plant leaves.*
  - b) *Mayer concluded that the disease was infectious when he found that he could transmit the disease by spraying sap from diseased leaves onto healthy plants.*
  - c) *He concluded that the disease must be caused by an extremely small bacterium, but Dimitri Ivanovsky demonstrated that the sap was still infectious even after passing through a filter designed to remove bacteria.*
2. In 1897 Martinus Beijerinck ruled out the possibility that the disease was due to a filterable toxin produced by a bacterium and demonstrated that the infectious agent could reproduce.
  - a) *The sap from one generation of infected plants could be used to infect a second generation of plants that could infect subsequent generations.*
  - b) *Beijerinck also determined that the pathogen could reproduce only within the host, could not be cultivated on nutrient media, and was not inactivated by alcohol, generally lethal to bacteria.*
3. In 1935, Wendell Stanley

### B. A virus is a genome enclosed in a protective coat

1. Stanley's discovery that some viruses could be crystallized was puzzling because not even the simplest cells can aggregate into regular crystals.
2. However, viruses are not cells.

3. They are infectious particles consisting of nucleic acid encased in a protein coat, and, in some cases, a membranous envelope.
4. Viruses range in size from only 20nm in diameter to that barely resolvable with a light microscope.
5. The genome of viruses includes other options than the double-stranded DNA that we have studied.
  - a) *Viral genomes may consist of double-stranded DNA, single-stranded DNA, double-stranded RNA, or single-stranded RNA, depending on the specific type of virus.*
  - b) *The viral genome is usually organized as a single linear or circular molecule of nucleic acid.*
  - c) *The smallest viruses have only four genes, while the largest have several hundred.*
6. The \_\_\_\_\_ is a protein shell enclosing the viral genome.
  - a) *Capsids are build of a large number of protein subunits called capsomeres, but with limited diversity.*
  - b) *The capsid of the tobacco mosaic virus has over 1,000 copies of the same protein.*
  - c) *Adenoviruses have 252 identical proteins arranged into a polyhedral capsid - as an icosahedron.*
  - d) *Some viruses have \_\_\_\_\_ , membranes cloaking their capsids.*
  - e) *These envelopes are derived from the membrane of the host cell.*
  - f) *They also have some viral proteins and glycoproteins.*
  - g) *The most complex capsids are found in viruses that infect bacteria, called \_\_\_\_\_ or \_\_\_\_\_ .*
  - h) *The T-even phages that infect Escherichia coli have a 20-sided capsid head that encloses their DNA and a protein tail piece that attaches the phage to the host and injects the phage DNA inside.*

### C. Viruses can reproduce only within a host cell: *an overview*

1. Viruses are obligate intracellular parasites.
2. An isolated virus is merely a packaged set of genes in transit from one host cell to another.
3. Each type of virus can infect and parasitize only a limited range of host cells, called its \_\_\_\_\_ .
4. Viruses identify host cells by a “lock-and-key” fit between proteins on the outside of virus and specific receptor molecules on the host’s surface.
5. Most viruses of eukaryotes attack specific tissues.
  - a) *Human cold viruses infect only the cells lining the upper respiratory tract.*
  - b) *The AIDS virus binds only to certain white blood cells.*
6. A viral infection begins when the genome of the virus enters the host cell.

7. Once inside, the viral genome commandeers its host, reprogramming the cell to copy viral nucleic acid and manufacture proteins from the viral genome.
8. The nucleic acid molecules and capsomeres then self-assemble into viral particles and exit the cell.

#### D. Phages reproduce using lytic or lysogenic cycles

1. While phages are the best understood of all viruses, some of them are also among the most complex.
2. Research on phages led to the discovery that some double-stranded DNA viruses can reproduce by two alternative mechanisms: the lytic cycle and the lysogenic cycle.

a) In the \_\_\_\_\_, \_\_\_\_\_, the phage reproductive cycle culminates in the death of the host.

- In the last stage, the bacterium lyses (breaks open) and releases the phages produced within the cell to infect others.
- \_\_\_\_\_ reproduce only by a lytic cycle.
- While phages have the potential to wipe out a bacterial colony in just hours, bacteria have defenses against phages.
- Natural selection favors bacterial mutants with receptors sites that are no longer recognized by a particular type of phage.
- Bacteria produce *restriction nucleases* that recognize and cut up foreign DNA, including certain phage DNA.
- Modifications to the bacteria's own DNA prevent its destruction by restriction nucleases.
- But, natural selection favors resistant phage mutants.

b) In the \_\_\_\_\_, \_\_\_\_\_, the phage genome replicates without destroying the host cell.

- \_\_\_\_\_, like phage lambda, use both lytic and lysogenic cycles.
- Within the host, the virus' circular DNA engages in either the lytic or lysogenic cycle.
- During a lytic cycle, the viral genes immediately turn the host cell into a virus-producing factory, and the cell soon lyses and releases its viral products.
- During the lysogenic cycle, the viral DNA molecule, is incorporated by genetic recombination into a specific site on the host cell's chromosome.
- In this \_\_\_\_\_ stage, one of its genes codes for a protein that represses most other prophage genes.
- Every time the host divides, it also copies the viral DNA and passes the copies to daughter cells.

- Occasionally, the viral genome exits the bacterial chromosome and initiates a lytic cycle.
- c) *This switch from lysogenic to lytic may be initiated by an environmental trigger.*
- d) *The lambda phage that infects E. coli demonstrates the cycles of a temperate phage.*

## E. Animal viruses are diverse in their modes of infection and replication

**Many variations on the basic scheme of viral infection and reproduction are represented among animal viruses.**

**One key variable is the type of nucleic acid that serves as a virus's genetic material.**

**Another variable is the presence or absence of a membranous envelope.**

1. Viruses equipped with an outer envelope use the envelope to enter the host cell.
  - a) *Glycoproteins on the envelope bind to specific receptors on the host's membrane.*
  - b) *The envelope fuses with the host's membrane, transporting the capsid and viral genome inside.*
  - c) *The viral genome duplicates and directs the host's protein synthesis machinery to synthesize capsomeres with free ribosomes and glycoproteins with bound ribosomes.*
  - d) *After the capsid and viral genome self-assemble, they bud from the host cell covered with an envelope derived from the host's plasma membrane, including viral glycoproteins.*
2. These enveloped viruses do not necessarily kill the host cell.
3. Some viruses have envelopes that are not derived from plasma membrane.
  - a) *The envelope of the herpesvirus is derived from the nuclear envelope of the host.*
    - These double-stranded DNA viruses reproduce within the cell nucleus using viral and cellular enzymes to replicate and transcribe their DNA.
    - Herpesvirus DNA may become integrated into the cell's genome as a provirus.
    - The \_\_\_\_\_ remains latent within the nucleus until triggered by physical or emotional stress to leave the genome and initiate active viral production.
4. The viruses that use RNA as the genetic material are quite diverse, especially those that infect animals.
  - In some with single-stranded RNA (class IV), the genome acts as mRNA and is translated directly.
  - In others (class V), the RNA genome serves as a *template* for mRNA and for a complementary RNA.
  - This complementary strand is the template for the synthesis of additional copies of genome RNA.
  - All viruses that require RNA → RNA synthesis to make mRNA use a viral enzyme that is packaged with the genome inside the capsid.
5. \_\_\_\_\_ (class VI) have the most complicated life cycles.

- a) These carry an enzyme, \_\_\_\_\_, which transcribes DNA from an RNA template.
- b) The newly made DNA is inserted as a provirus into a chromosome in the animal cell.
- c) The host's RNA polymerase transcribes the viral DNA into more RNA molecules.
- These can function both as mRNA for the synthesis of viral proteins and as genomes for new virus particles released from the cell.
6. Human immunodeficiency virus (HIV), the virus that causes AIDS (acquired immunodeficiency syndrome) is a retrovirus.
- a) The viral particle includes an envelope with glycoproteins for binding to specific types of red blood cells, a capsid containing two identical RNA strands as its genome and two copies of reverse transcriptase.
- b) The reproductive cycle of HIV illustrates the pattern of infection and replication in a retrovirus.
- c) After HIV enters the host cell, reverse transcriptase synthesizes double stranded DNA from the viral RNA.
- d) Transcription produces more copies of the viral RNA that are translated into viral proteins, which self-assemble into a virus particle and leave the host.
7. The link between viral infection and the symptoms it produces is often obscure.
- a) Some viruses damage or kill cells by triggering the release of hydrolytic enzymes from lysosomes.
- b) Some viruses cause the infected cell to produce toxins that lead to disease symptoms.
- c) Others have molecular components, such as envelope proteins, that are toxic.
8. In some cases, viral damage is easily repaired (respiratory epithelium after a cold), but in others, infection causes permanent damage (nerve cells after polio).
9. Many of the temporary symptoms associated with a viral infection results from the body's own efforts at defending itself against infection.
- a) The immune system is a complex and critical part of the body's natural defense mechanism against viral and other infections.
10. Modern medicine has developed \_\_\_\_\_, harmless variants or derivatives of pathogenic microbes, which stimulate the immune system to mount defenses against the actual pathogen.
- a) The first vaccine was developed in the late 1700s by Edward Jenner to fight smallpox.
- Jenner learned from his patients that milkmaids who had contracted cowpox, a milder disease that usually infects cows, were resistant to smallpox.
  - In his famous experiment in 1796, Jenner infected a farm boy with cowpox, acquired from the sore of a milkmaid with the disease.
  - When exposed to smallpox, the boy resisted the disease.
  - Because of their similarities, vaccination with the cowpox virus sensitizes the immune system to react vigorously if exposed to actual smallpox virus.

- b) *Effective vaccines against many other viruses exist.*
- c) *Vaccines can help prevent viral infections, but they can do little to cure most viral infection once they occur.*
- d) *Antibiotics, which can kill bacteria by inhibiting enzymes or processes specific to bacteria, are powerless against viruses, which have few or no enzymes of their own.*
- e) *Some recently developed drugs do combat some viruses, mostly by interfering with viral nucleic acid synthesis.*
  - AZT interferes with reverse transcriptase of HIV.
  - Acyclovir inhibits herpesvirus DNA synthesis.

11. In recent years, several very dangerous “emergent viruses” have risen to prominence.

- HIV, the AIDS virus, seemed to appear suddenly in the early 1980s.
  - Each year new strains of influenza virus cause millions to miss work or class, and deaths are not uncommon.
  - The deadly Ebola virus has caused hemorrhagic fevers in central Africa periodically since 1976.
- a) *The emergence of these new viral diseases is due to three processes: mutation, spread of existing viruses from one species to another, and dissemination of a viral disease from a small, isolated population.*
- b) *Mutation of existing viruses is a major source of new viral diseases.*
- RNA viruses tend to have high mutation rates because replication of their nucleic acid lacks proofreading.
  - Some mutations create new viral strains with sufficient genetic differences from earlier strains that they can infect individuals who had acquired immunity to these earlier strains.
    - This is the case in flu *epidemics*.
- c) *Another source of new viral diseases is the spread of existing viruses from one host species to another.*
- It is estimated that about three-quarters of new human diseases have originated in other animals.
  - For example, hantavirus, which killed dozens of people in 1993, normally infects rodents, especially deer mice.
  - That year unusually wet weather in the southwestern U.S. increased the mice’s food, exploding its population.
  - Humans acquired hantavirus when they inhaled dust containing traces of urine and feces from infected mice.
- d) *Finally, a viral disease can spread from a small, isolated population to a widespread epidemic.*
- For example, AIDS went unnamed and virtually unnoticed for decades before spreading around the world.
  - Technological and social factors, including affordable international travel, blood transfusion technology, sexual promiscuity, and the abuse of intravenous drugs, allowed a previously rare disease to become a global scourge.

- These emerging viruses are generally not new. Rather, they are existing viruses that expand their host territory.
- Environmental change can increase the viral traffic responsible for emerging disease.
- Since 1911, when Peyton Rous discovered that a virus causes cancer in chickens, scientists have recognized that some viruses cause animal cancers.
  - These *tumor viruses* include retrovirus, papovavirus, adenovirus, and herpesvirus types.

12. Viruses appear to cause certain human cancers.

- The hepatitis B virus is associated with liver cancer.*
- The Epstein-Barr virus, which causes infectious mononucleosis, has been linked to several types of cancer in parts of Africa, notably Burkitt's lymphoma.*
- Papilloma viruses are associated with cervical cancers.*
- The HTLV-1 retrovirus causes a type of adult leukemia.*

13. All tumor viruses transform cells into cancer cells after integration of viral nucleic acid into host DNA.

- Viruses may carry oncogenes that trigger cancerous characteristics in cells.*
  - These oncogenes are often versions of *proto-oncogenes* that influence the cell cycle in normal cells.
  - Proto-oncogenes generally code for growth factors or proteins involved in growth factor function.
- In other cases, a tumor virus transforms a cell by turning on or increasing the expression of proto-oncogenes.*
- It is likely that most tumor viruses cause cancer only in combination with other mutagenic events.*

## F. Plant viruses are serious agricultural pests

- Plant viruses can stunt plant growth and diminish crop yields.
- Most are RNA viruses with rod-shaped capsids produced by a spiral of capsomeres.
- Plant viral diseases are spread by two major routes.
- In *horizontal transmission*, a plant is infected with the virus by an external source.
  - Plants are more susceptible if their protective epidermis is damaged, perhaps by wind, chilling, injury, or insects.*
  - Insects are often carriers of viruses, transmitting disease from plant to plant.*
- In *vertical transmission*, a plant inherits a viral infection from a parent.
  - This may occur by asexual propagation or in sexual reproduction via infected seeds.*
- Once it starts reproducing inside a plant cell, virus particles can spread throughout the plant by passing through plasmodesmata.

- a) *These cytoplasmic connections penetrate the walls between adjacent cells.*
7. Agricultural scientists have focused their efforts largely on reducing the incidence and transmission of viral disease and in breeding resistant plant varieties.

### G. Viroids and prions are infectious agents even simpler than viruses

1. \_\_\_\_\_, smaller and simpler than even viruses, consist of tiny molecules of naked circular RNA that infect plants.
- a) *Their several hundred nucleotides do not encode for proteins but can be replicated by the host's cellular enzymes.*
- b) *These RNA molecules can disrupt plant metabolism and stunt plant growth, perhaps by causing errors in the regulatory systems that control plant growth.*
2. \_\_\_\_\_ are infectious *proteins* that spread a disease.
- a) *They appear to cause several degenerative brain diseases including scrapie in sheep, "mad cow disease," and Creutzfeldt-Jacob disease in humans.*
- b) *According to the leading hypothesis, a prion is a misfolded form of a normal brain protein.*
- c) *It can then convert a normal protein into the prion version, creating a chain reaction that increases their numbers.*

### H. Viruses may have evolved from other mobile genetic elements

**Because viruses depend on cells for their own propagation, it is reasonable to assume that they evolved after the first cells appeared.**

1. Most molecular biologists favor the hypothesis that viruses originated from fragments of cellular nucleic acids that could move from one cell to another.
- a) *A viral genome usually has more in common with the genome of its host than with those of viruses infecting other hosts.*
- b) *Perhaps the earliest viruses were naked bits of nucleic acids that passed between cells via injured cell surfaces.*
- c) *The evolution of capsid genes may have facilitated the infection of undamaged cells.*
2. Candidates for the original sources of viral genomes include plasmids and transposons.
- a) *Plasmids are small, circular DNA molecules that are separate from chromosomes.*
- b) *Plasmids, found in bacteria and in the eukaryote yeast, can replicate independently of the rest of the cell and are occasionally transferred between cells.*
- c) *Transposons are DNA segments that can move from one location to another within a cell's genome.*
- d) *Both plasmids and transposons are mobile genetic elements.*

### III. The Genetics of Bacteria

#### A. The short generation span of bacteria helps them adapt to changing environments

- a) *This is true in the evolutionary sense of adaptation via natural selection and the physiological sense of adjustment to changes in the environment by individual bacteria.*
2. The major component of the bacterial genome is one double-stranded, circular DNA molecule.
  - a) *For E. coli, the chromosomal DNA consists of about 4.6 million nucleotide pairs with about 4,300 genes.*
  - b) *This is 100 times more DNA than in a typical virus and 1,000 times less than in a typical eukaryote cell.*
  - c) *Tight coiling of the DNA results in a dense region of DNA, called the \_\_\_\_\_, not bound by a membrane.*
3. In addition, many bacteria have \_\_\_\_\_, much smaller circles of DNA.
  - a) *Each plasmid has only a small number of genes, from just a few to several dozen.*
4. Bacteria proliferate very rapidly in a favorable natural or laboratory environment.
  - a) *Under optimal laboratory conditions E. coli can divide every 20 minutes, producing a colony of  $10^7$  to  $10^8$  bacteria in as little as 12 hours.*
  - b) *In the human colon, E. coli reproduces rapidly enough to replace the  $2 \times 10^{10}$  bacteria lost each day in feces.*
5. Through binary fission, most of the bacteria in a colony are genetically identical to the parent cell.
  - a) *However, the spontaneous mutation rate of E. coli is  $1 \times 10^{-7}$  mutations per gene per cell division.*
  - b) *This will produce about 2,000 bacteria in the human colon that have a mutation in that gene per day.*
6. New mutations, though individually rare, can have a significant impact on genetic diversity when reproductive rates are very high because of short generation spans.
7. Individual bacteria that are genetically well equipped for the local environment clone themselves more prolifically than do less fit individuals.
8. In contrast, organisms with slower reproduction rates (like humans) create most genetic variation not by novel alleles produced through mutation, but by sexual recombination of existing alleles.

#### B. Genetic recombination produces new bacterial strains

1. In addition to mutations, genetic recombination generates diversity within bacterial populations.
2. Here, recombination is defined as the combining of DNA from two individuals into a single genome.

- a) *Recombination occurs through three processes:*
- Transformation
  - Transduction
  - Conjugation
- b) *The impact of recombination can be observed when two mutant E. coli strains are combined.*
- c) *If each is unable to synthesize one of its required amino acids, neither can grow on a minimal medium.*
- d) *However, if they are combined, numerous colonies will be created that started as cells that acquired the missing genes for amino acid synthesis from the other strain.*
- e) *Some may have resulted from mutation.*
3. \_\_\_\_\_ is the alteration of a bacterial cell's genotype by the uptake of naked, foreign DNA from the surrounding environment.
- a) *For example, harmless Streptococcus pneumoniae bacteria can be transformed to pneumonia-causing cells.*
- b) *This occurs when a live nonpathogenic cell takes up a piece of DNA that happens to include the allele for pathogenicity from dead, broken-open pathogenic cells.*
- c) *The foreign allele replaces the native allele in the bacterial chromosome by genetic recombination.*
- d) *The resulting cell is now recombinant with DNA derived from two different cells.*
4. Many bacterial species have surface proteins that are specialized for the uptake of naked DNA.
- a) *These proteins recognize and transport only DNA from closely related bacterial species.*
- b) *While E. coli lacks this specialized mechanism, it can be induced to take up small pieces of DNA if cultured in a medium with a relatively high concentration of calcium ions.*
- c) *In biotechnology, this technique has been used to introduce foreign DNA into E. coli.*
5. \_\_\_\_\_ occurs when a phage carries bacterial genes from one host cell to another.
6. In \_\_\_\_\_, a small piece of the host cell's degraded DNA is packaged within a capsid, rather than the phage genome.
- a) *When this phage attaches to another bacterium, it will inject this foreign DNA into its new host.*
- b) *Some of this DNA can subsequently replace the homologous region of the second cell.*
- c) *This type of transduction transfers bacterial genes at random.*
7. \_\_\_\_\_ occurs via a temperate phage.

- a) *When the prophage viral genome is excised from the chromosome, it sometimes takes with it a small region of adjacent bacterial DNA.*
  - b) *These bacterial genes are injected along with the phage's genome into the next host cell.*
  - c) *Specialized transduction only transfers those genes near the prophage site on the bacterial chromosome.*
8. Both generalized and specialized transduction use phage as a vector to transfer genes between bacteria.
  9. \_\_\_\_\_ transfers genetic material between two bacterial cells that are temporarily joined.
  10. One cell ("male") donates DNA and its "mate" ("female") receives the genes.
  11. A sex pilus from the male initially joins the two cells and creates a cytoplasmic bridge between cells.
  12. "Maleness," the ability to form a sex pilus and donate DNA, results from an \_\_\_\_\_ as a section of the bacterial chromosome or as a plasmid.
  13. \_\_\_\_\_, including the F plasmid, are small, circular, self-replicating DNA molecules.
  14. \_\_\_\_\_, like the F plasmid, can undergo reversible incorporation into the cell's chromosome.
    - a) *Temperate viruses also qualify as episomes.*
  15. Plasmids generally benefit the bacterial cell.
  16. They usually have only a few genes that are not required for normal survival and reproduction.
    - a) *Plasmid genes are advantageous in stressful conditions.*
      - The F plasmid facilitates genetic recombination when environmental conditions no longer favor existing strains.
  17. The F factor or its \_\_\_\_\_ consists of about 25 genes, most required for the production of sex pili.
    - a) *Cells with either the F factor or the F plasmid are called F<sup>+</sup> and they pass this condition to their offspring.*
    - b) *Cells lacking either form of the F factor, are called F<sup>-</sup>, and they function as DNA recipients.*
  18. When an F<sup>+</sup> and F<sup>-</sup> cell meet, the F<sup>+</sup> cell passes a copy of the F plasmid to the F<sup>-</sup> cell, converting it.
  19. The plasmid form of the F factor can become integrated into the bacterial chromosome.

20. The resulting Hfr cell (high frequency of recombination) functions as a male during conjugation.
21. The Hfr cell initiates DNA replication at a point on the F factor DNA and begins to transfer the DNA copy from that point to its F<sup>-</sup> partner
22. Random movements almost always disrupt conjugation long before an entire copy of the Hfr chromosome can be passed to the F<sup>-</sup> cell.
23. In the partially diploid cell, the newly acquired DNA aligns with the homologous region of the F<sup>-</sup> chromosome.
24. Recombination exchanges segments of DNA.
25. This recombinant bacteria has genes from two different cells.
26. In the 1950s, Japanese physicians began to notice that some bacterial strains had evolved antibiotic resistance.
  - a) *The genes conferring resistance are carried by plasmids, specifically the \_\_\_\_\_ (R for resistance).*
  - b) *Some of these genes code for enzymes that specifically destroy certain antibiotics, like tetracycline or ampicillin.*
27. When a bacterial population is exposed to an antibiotic, individuals with the R plasmid will survive and increase in the overall population.
28. Because R plasmids also have genes that encode for sex pili, they can be transferred from one cell to another by conjugation.
29. A \_\_\_\_\_ is a piece of DNA that can move from one location to another in a cell's genome.
30. Transposon movement occurs as a type of recombination between the transposon and another DNA site, a target site.
  - a) *In bacteria, the target site may be within the chromosome, from a plasmid to chromosome (or vice versa), or between plasmids.*
31. Transposons can bring multiple copies for antibiotic resistance into a single R plasmid by moving genes to that location from different plasmids.
  - a) *This explains why some R plasmids convey resistance to many antibiotics.*
32. Some transposons (so called "jumping genes") do jump from one location to another (cut-and-paste translocation).
33. However, in replicative transposition, the transposon replicates at its original site, and a copy inserts elsewhere.
34. Most transposons can move to many alternative locations in the DNA, potentially moving genes to a site where genes of that sort have never before existed.

35. The simplest bacterial transposon, an \_\_\_\_\_ , consists only of the DNA necessary for the act of transposition.
36. The insertion sequence consists of the transposase gene, flanked by a pair of *inverted repeat* sequences.
- a) *The 20 to 40 nucleotides of the inverted repeat on one side are repeated in reverse along the opposite DNA strand at the other end of the transposon.*
37. The transposase enzyme recognizes the inverted repeats as the edges of the transposon.
38. Transposase cuts the transposon from its initial site and inserts it into the target site.
- a) *Gaps in the DNA strands are filled in by DNA polymerase, creating direct repeats, and then DNA ligase seals the old and new material.*
39. Insertion sequences cause mutations when they happen to land within the coding sequence of a gene or within a DNA region that regulates gene expression.
40. Insertion sequences account for 1.5% of the *E. coli* genome, but a mutation in a particular gene by transposition is rare, about 1 in every 10 million generations.
- a) *This is about the same rate as spontaneous mutations from external factors.*
41. *Composite transposons* (complex transposons) include extra genes sandwiched between two insertion sequences.
- a) *It is as though two insertion sequences happened to land relatively close together and now travel together, along with all the DNA between them, as a single transposon.*
42. While insertion sequences may not benefit bacteria in any specific way, composite transposons may help bacteria adapt to new environments.
- a) *For example, repeated movements of resistance genes by composite transposition may concentrate several genes for antibiotic resistance onto a single R plasmid.*
- b) *In an antibiotic-rich environment, natural selection favors bacterial clones that have built up composite R plasmids through a series of transpositions.*
43. Transposable genetic elements are important components of eukaryotic genomes as well.
44. In the 1940s and 1950s Barbara McClintock investigated changes in the color of corn kernels.
- a) *She postulated that the changes in kernel color only made sense if mobile genetic elements moved from other locations in the genome to the genes for kernel color.*
- b) *When these “controlling elements” were inserted next to the genes responsible for kernel color, they would activate or inactivate those genes.*
- c) *In 1983, more than 30 years after her initial break-through, Dr. McClintock received a Nobel Prize for her discovery.*

C. The control of gene expression enables individual bacteria to adjust their metabolism to environmental change

1. An individual bacterium, locked into the genome that it has inherited, can cope with environmental fluctuations by exerting metabolic control.
  - a) *First, cells vary the number of specific enzyme molecules by regulating gene expression.*
  - b) *Second, cells adjust the activity of enzymes already present (for example, by feedback inhibition).*
2. For example, the tryptophan biosynthesis pathway demonstrates both levels of control.
  - a) *If tryptophan levels are high, some of the tryptophan molecules can inhibit the first enzyme in the pathway.*
  - b) *If the abundance of tryptophan continues, the cell can stop synthesizing additional enzymes in this pathway by blocking transcription of the genes for these enzymes.*
3. In 1961, Francois Jacob and Jacques Monod proposed the operon model for the control of gene expression in bacteria.
4. An \_\_\_\_\_ consists of three elements:
  - a) *The genes that it controls.*
    - In bacteria, the genes coding for the enzymes of a particular pathway are clustered together and transcribed (or not) as one long mRNA molecule.
  - b) *A promotor region where RNA polymerase first binds.*
  - c) *An \_\_\_\_\_ region between the promotor and the first gene that acts as an “on-off switch.”*
5. By itself, an operon is on and RNA polymerase can bind to the promotor and transcribe the genes.
6. However, if a \_\_\_\_\_ protein, a product of a \_\_\_\_\_, binds to the operator, it can prevent transcription of the operon's genes.
  - a) *Each repressor protein recognizes and binds only to the operator of a certain operon.*
  - b) *Regulatory genes are transcribed continuously at low rates.*
7. Binding by the repressor to the operator is reversible.
  - a) *The number of active repressor molecules available determines the on and off mode of the operator.*
8. Many repressors contain allosteric sites that change shape depending on the binding of other molecules.

- a) In the case of the *trp* operon, when concentrations of tryptophan in the cell are high, some tryptophan molecules bind as a \_\_\_\_\_ to the repressor protein.
- b) This activates the repressor and turns the operon off.
- c) At low levels of tryptophan, most of the repressors are inactive and the operon is transcribed.
9. The *trp* operon is an example of a *repressible* operon, one that is *inhibited* when a specific small molecule binds allosterically to a regulatory protein.
10. In contrast, an *inducible* operon is *stimulated* when a specific small molecule interacts with a regulatory protein.
- a) In inducible operons, the regulatory protein is active (inhibitory) as synthesized, and the operon is off.
- b) Allosteric binding by an \_\_\_\_\_ molecule makes the regulatory protein inactive, and the operon is on.
11. The *lac* operon contains a series of genes that code for enzymes that play a major role in the hydrolysis and metabolism of lactose.
- a) In the absence of lactose, this operon is off as an active repressor binds to the operator and prevents transcription.
12. When lactose is present in the cell, allolactose, an isomer of lactose, binds to the repressor.
- a) This inactivates the repressor, and the *lac* operon can be transcribed.
13. Repressible enzymes generally function in anabolic pathways, synthesizing end products.
- a) When the end product is present in sufficient quantities, the cell can allocate its resources to other uses.
14. Inducible enzymes usually function in catabolic pathways, digesting nutrients to simpler molecules.
- a) By producing the appropriate enzymes only when the nutrient is available, the cell avoids making proteins that have nothing to do.
15. Both repressible and inducible operons demonstrate *negative* control because active repressors can only have negative effects on transcription.
16. Positive gene control occurs when an activator molecule interacts directly with the genome to switch transcription on.
17. Even if the *lac* operon is turned on by the presence of allolactose, the degree of transcription depends on the concentrations of other substrates.
- a) If glucose levels are low (along with overall energy levels), then \_\_\_\_\_  
 \_\_\_\_\_ (**cAMP**) binds to **cAMP** \_\_\_\_\_  
 \_\_\_\_\_ (**CRP**) which activates transcription.
18. The cellular metabolism is biased toward the utilization of glucose.

19. If glucose levels are sufficient and cAMP levels are low (lots of ATP), then the CRP protein has an inactive shape and cannot bind upstream of the *lac* promoter.
  - a) *The lac operon will be transcribed but at a low level.*
20. For the *lac* operon, the presence / absence of lactose (allolactose) determines if the operon is on or off.
21. Overall energy levels in the cell determine the level of transcription, a “volume” control, through CRP.
22. CRP works on several operons that encode enzymes used in catabolic pathways.
  - a) *If glucose is present and CRP is inactive, then the synthesis of enzymes that catabolize other compounds is slowed.*
  - b) *If glucose levels are low and CRP is active, then the genes which produce enzymes that catabolize whichever other fuel is present will be transcribed at high levels.*