13 Characterizing and Classifying Viruses, Viroids, and Prions

CHAPTER SUMMARY

Viruses, viroids, and prions are **acellular** (noncellular) disease-causing agents that lack cell structure and cannot metabolize, grow, reproduce, or respond to their environment. They must recruit the cell's metabolic chemicals and ribosomes in order to increase their numbers.

Characteristics of Viruses (pp. 378–382)

A **virus** is a miniscule, acellular, infectious agent having one or several pieces of nucleic acid—either DNA or RNA, but never both. Viruses have no cytoplasmic membrane, and with one exception lack organelles and cytosol. In its **extracellular state**, a virus is called a **virion**. It consists of a protein coat, called a **capsid**, surrounding a nucleic acid core. Together the nucleic acid and its capsid are called a *nucleocapsid*. Some virions have a phospholipid membrane called an *envelope* surrounding the nucleocapsid. When a virus penetrates a cell, the **intracellular state** is initiated; the capsid is removed. A virus without a capsid exists solely as nucleic acid but is still referred to as a virus.

Genetic Material of Viruses

The genome of viruses includes either DNA or RNA, but never both. In addition, they may be double-stranded (ds) or single-stranded (ss). Thus, viral genomes are described as dsDNA, ssDNA, dsRNA, or ssRNA. They may exist as multiple linear molecules of nucleic acid, or circular and singular molecules of nucleic acid, depending on the type of virus. Viral genomes are usually much smaller than the genomes of cells. The smallest chlamydial bacterium has almost 1000 genes; the genome of bacteriophage MSZ has only three genes.

Hosts of Viruses

Most viruses infect only particular kinds of cells. This specificity is due to the affinity of viral surface proteins or glycoproteins for complementary proteins or glycoproteins on the surface of the host cell. A virus that infects bacteria is referred to as a **bacteriophage**, or simply a **phage**. Viruses also infect humans, other animals, plants, and even fungi.

Sizes of Viruses

Viruses are so small that most cannot be seen by light microscopy. The smallest have a diameter of 10 nm, whereas the largest are approximately 400 nm, about the size of the smallest bacterial cell.

Capsid Morphology

The capsid of a virus is composed of proteinaceous subunits called **capsomeres**. These may be composed of only a single type of protein, or of several different protein molecules.

Viral Shapes

There are three basic types of viral shapes: helical viruses have capsomeres that spiral around the nucleic acid, forming a tube-like structure; polyhedral viruses are roughly spherical, with a shape similar to a geodesic dome; and complex viruses have capsids of many different shapes.

The Viral Envelope

Some viruses have a membrane similar in composition to a cell membrane surrounding their capsids. This membrane is called an **envelope**, and thus a virus with a membrane is called an *enveloped virion*. A virion without an envelope is a *nonenveloped* or *naked virion*. The envelope of a virus is acquired from the host cell during replication or release, and is a portion of the host cell's membrane system.

Classification of Viruses (pp. 382–385)

Virologists classify viruses by their type of nucleic acid, presence of an envelope, shape, and size. They have recognized viral family and genus names. With the exception of three orders, higher taxa are not established. At this time, specific epithets for viruses are their common English designations written in italics, such as *rabies virus*.

Viral Replication (pp. 385–394)

Viruses cannot reproduce themselves because they have neither the genes for all enzymes necessary for replication nor do they possess functional ribosomes for protein synthesis. Instead, they depend on random contact with a specific host cell type for the organelles and enzymes to produce new virions.

Lytic Replication of Bacteriophages

Viral replication that results in lysis of the cell near the end of the cycle is termed **lytic replication**. The cycle consists of five stages:

1. During **attachment**, the virion attaches to the host cell.
2. During **entry**, the virion or its genome enters the host cell. In bacteriophages, only the nucleic acid enters the cell.
3. During **synthesis**, the host cell's metabolic enzymes and ribosomes are used to synthesize new nucleic acids and viral proteins.
4. During **assembly**, new virions are spontaneously assembled in the host cell, typically as capsomeres surround replicated or transcribed nucleic acids to form new virions.
5. During **release**, new virions are released from the host cell, which lyses.

Lysogenic Replication of Bacteriophages

Not all viruses follow the lytic pattern. Some bacteriophages have a modified replication cycle in which infected host cells grow and reproduce normally for many generations before they lyse. Such a replication cycle is called a **lysogenic replication cycle** or **lysogeny**, and the phages involved are called **lysogenic phages** or **temperate phages**. After entry into the host cell, the viral genome does not immediately assume control of the cell but instead remains inactive. Such an inactive phage is called a **prophage**. A prophage is always inserted into the DNA of the bacterium, becomes

a physical part of the bacterial chromosome, and is passed on to daughter cells. Lysogenic phages can change the phenotype of a bacterium by the process of lysogenic conversion. At some point in the generations that follow, a prophage may be excised from the chromosome in a process known as **induction**. At that point, the prophage again becomes a lytic virus.

Replication of Animal Viruses

Animal viruses have the same basic replication pathway as bacteriophages, but some differences result in part from the presence of envelopes around some of the viruses and in part from the eukaryotic nature of animal cells and their lack of a cell wall.

Attachment of Animal Viruses

Animal viruses lack both tails and tail fibers, and typically attach via glycoprotein spikes or other molecules on their capsids or envelopes.

Entry and Uncoating of Animal Viruses

Some naked viruses enter their host's cells by direct penetration, a process in which a viral capsid attaches and sinks into the cytoplasmic membrane. This creates a pore through which the viral genome alone enters the cell. In enveloped viruses, the entire capsid enters the cell. In some viruses, the viral envelope and host cell membrane fuse, releasing the capsid into the cell's cytoplasm and leaving the envelope glycoproteins as part of the cell membrane. In other cases, the entire virus is endocytized, and the capsid must be removed to release the genome. This removal of a viral capsid within a host cell is called **uncoating**.

Synthesis of Animal Viruses

Each type of animal virus requires a different strategy for synthesis that depends on the kind of nucleic acid involved: DNA or RNA, and ds versus ss:

- Synthesis of new dsDNA virions is similar to the normal replication of cellular DNA and translation of proteins. Each strand of viral DNA is used as a template for its complement. This method of replication is seen with herpes and papilloma viruses; in poxviruses, synthesis occurs in the cytoplasm.
- Parvovirus, a human virus with ssDNA, is synthesized by host cell enzymes, which synthesize a complement to the ssDNA. The complementary strand binds to the ssDNA of the virus to form a dsDNA molecule. Transcription, replication, and assembly then follow.
- Some ssRNA viruses have **positive strand RNA** (+ssRNA), which can be directly translated by ribosomes to synthesize protein. From the +ssRNA, complementary **negative strand RNA** (–ssRNA) is also transcribed to serve as a template for more +ssRNA. **Retroviruses** such as HIV are +ssRNA viruses that carry *reverse transcriptase* to transcribe DNA from their RNA. This reverse process (DNA transcribed from RNA) is reflected in the name retrovirus.
- Viruses with –ssRNA carry an RNA-dependent RNA transcriptase for transcribing mRNA from the –ssRNA genome so that protein can then be translated. Transcription of RNA from RNA is not found in cells.
- When dsRNA functions as the genome of some viruses, one strand of the RNA molecule functions as the genome, and the other strand functions as a template for RNA replication.

Assembly and Release of Animal Viruses

Enveloped animal viruses are often released via a process called **budding**. As virions are assembled, they are extruded through one of the cell's membranes—the nuclear membrane, the endoplasmic reticulum, or the cell membrane. Each virion acquires a portion of cell membrane, which becomes its envelope. Budding allows an infected cell to remain alive for some time.

Latency of Animal Viruses

Some animal viruses, including chickenpox and herpesviruses, may remain dormant in cells in a process known as **latency**; the viruses involved are called **latent viruses** or **proviruses**. Latency may be prolonged for years. Unlike a lysogenic bacteriophage, a provirus does not typically become incorporated into the host cell's chromosomes. When it does so, as with HIV, the provirus remains there permanently; induction never occurs.

The Role of Viruses in Cancer (pp. 394–396)

Neoplasia is uncontrolled cellular reproduction in a multicellular animal. A mass of neoplastic cells, called a **tumor**, may be relatively harmless (**benign**) or invasive (**malignant**). Malignant tumors are also called **cancer**. The term **metastasis** describes the spreading of malignant cells, which rob normal cells of space and nutrients, cause pain, and derange the function of affected tissues until eventually the body can no longer withstand the loss of normal function and dies.

Several theories have been proposed to explain the role viruses play in cancer. These theories revolve around the presence of *proto-oncogenes* that play a role in cell division. Viruses have been implicated as possible activators of oncogenes (activated proto-oncogenes) and as inhibitors of oncogene repressors. Among known virally induced cancers in humans are Hodgkin's disease, Kaposi's sarcoma, cervical cancer, and others.

Culturing Viruses in the Laboratory (pp. 396–398)

Because viruses cannot metabolize or replicate by themselves, they cannot be grown in standard broths or on agar plates. Instead, they must be cultured inside suitable host cells.

Culturing Viruses in Whole Organisms

Most of our knowledge of viral replication has been derived from research on bacteriophages, which are relatively easy to culture because bacteria are easily grown and maintained. Phages can be grown in bacteria maintained in either liquid cultures or on agar plates. On plates, clear zones called **plaques** are areas where phages have lysed bacteria. Such plates enable the estimation of phage numbers via a technique called **plaque assay**.

Maintaining laboratory animals can be difficult and expensive, and the practice raises ethical concerns for some. Growing viruses that infect only humans raises additional ethical complications. Therefore, scientists have developed alternative ways of culturing animal and human viruses using fertilized chicken eggs or cell cultures.

Culturing Viruses in Embryonated Chicken Eggs

Chicken eggs are a useful culture medium for viruses because they are inexpensive, among the largest of cells, free of contaminating microbes, and contain a nourishing yolk. Most suitable are eggs that have been fertilized.

Culturing Viruses in Cell (Tissue) Culture

Viruses can also be grown in **cell culture**, which consists of cells isolated from an organism and grown on the surface of a medium or in broth. They are of two types: **Diploid cell cultures** are created from embryonic animal, plant, or human cells that have been isolated and provided appropriate growth conditions. **Continuous cell cultures** are more long lasting because they are derived from tumor cells, which divide relentlessly.

Other Parasitic Particles: Viroids and Prions (pp. 398–401)

Two molecular particles also infect cells: viroids and prions.

Characteristics of Viroids

Viroids are small, circular pieces of RNA with no capsid that infect and cause disease in plants. Similar RNA molecules affect some fungi.

Characteristics of Prions

Prions are infectious protein particles that lack nucleic acids and replicate by converting similar normal proteins into new prions. Diseases associated with prions are spongiform encephalopathies such as bovine spongiform encephalitis (BSE, so-called "mad cow disease"), scrapie in sheep, and Creutzfeldt-Jakob disease in humans. All of these involve fatal neurological degeneration. Prions are not destroyed by normal cooking or sterilization, though they are destroyed by incineration. There is no treatment for any prion disease.

Are Viruses Alive? (p. 401)

The characteristics of life are growth, self-reproduction, responsiveness, and the ability to metabolize. According to these criteria, viruses seem to lack the qualities of living things. For some scientists, however, three observations indicate that viruses are the least complex living entities: First, viruses use sophisticated methods to invade cells. Second, they take control of host cells. Third, they possess genomes containing the instructions for their own replication. Thus, viruses teeter on the threshold of life: Outside of cells, they do not appear to be alive, but within cells, they direct the synthesis and assembly required to make copies of themselves.

KEY THEMES

Chapter 13 is the last of the three chapters on essential characteristics and general classification schemes for the various microbial groups, and is the least precise. Viruses, viroids, and prions by their very nature defy classification in the tradition sense of biology. Remember the following while studying this chapter:

- *Viruses, viroids, and prions are acellular and do not form branches of the tree of life:* It is doubtful if any of these microscopic entities are living, and any relationships they may have had with living cells is so remote as to be impossible to uncover. When studying viruses, viroids, and prions, we are therefore studying microbes completely different from any of the other microbes we have come to know.

QUESTIONS FOR FURTHER REVIEW

Answers to these questions can be found in the answer section at the back of this study guide. Refer to the answers only after you have attempted to solve the questions on your own.

Multiple Choice

1. Which of the following is a true statement about viruses?
   a. Viruses are self-replicating
   b. Viruses are active outside of cells
   c. Viruses contain either DNA or RNA, but not both
   d. Viruses are capable of growth
2. Which of the following genome types is not found among viruses?
   a. dsDNA
   b. dsRNA
   c. ssRNA
   d. All of these are found among viruses
3. Phages are viruses that specifically infect:
   a. Algae
   b. Bacteria
   c. Fungi
   d. Protozoa
4. Naked virions lack:
   a. A genome
   b. A capsid
   c. An envelope
   d. All of the above
5. Which step of the viral replication cycle of T4 phage is performed for the virus by *E. coli*?
   a. Attachment
   b. Penetration
   c. Assembly
   d. All are performed by *E. coli*
6. Which event below generally occurs as a discrete step in the replication cycle of animal viruses but not in that of phages?
   a. Attachment
   b. Penetration
   c. Uncoating
   d. Assembly
7. Which method of penetration can be done by both phages and naked animal viruses?
   a. Direct injection of the genome across the cell membrane
   b. Fusion of the capsid with the cell membrane
   c. Stimulation of phagocytosis to bring the entire capsid inside
   d. None of the above are used by both
8. Which type of viral genome most closely resembles eukaryotic mRNA?
   a. dsDNA
   b. +ssRNA
   c. –ssRNA
   d. dsRNA

9. Can an RNA virus ever integrate into a host cell genome as a provirus?
   a. No, only DNA viruses can integrate
   b. Yes, any RNA virus can integrate
   c. Yes, but only if the virus is a retrovirus and has reverse transcriptase
   d. Proviruses never integrate into the host genome
10. How can a virus cause cancer?
    a. By introducing an oncogene
    b. By stimulating oncogenes that are already present
    c. By interfering with tumor repression
    d. All of the above
11. When studying a human virus in the laboratory, the best way to culture the virus would be to use:
    a. Bacteria
    b. Chicken eggs
    c. Mice
    d. Human cell culture
12. Viroids differ from viruses in that viroids:
    a. Are larger than viruses
    b. Are always linear whereas viruses are not
    c. Lack a capsid whereas viruses always have a capsid
    d. Infect only animals whereas viruses can infect any cell type
13. Prions are:
    a. Viruses
    b. Viroids
    c. Infectious proteins
    d. None of the above
14. Which of the following mechanisms is not a method by which prions are transmitted?
    a. Ingestion of infected tissue
    b. Transplantion of infected tissue
    c. Mucous membrane contact with infected tissue
    d. All of the above are methods of transmission

Fill in the Blanks

1. A virion is the _____ (intracellular/extracellular) state of a virus. The virion consists at a minimum of a protein _____ and a nucleic acid _____.
2. Viral capsids are constructed from protein subunits called _____.
3. The three capsid types seen among viruses are _____, _____, and _____.
4. Phage T4 releases an enzyme called _____ to aid in penetration of the host cell. This enzyme is also used during _____ (name the stage of replication).

5. Phages can undergo one of two types of replication inside a host cell, _____ or _____.
6. Though dsDNA viruses, _____ do not replicate in the nucleus as do other dsDNA viruses.
7. _____ are single-stranded DNA viruses.
8. HIV is a _____ (name the type of virus) that has a _____ genome. It carries the enzyme _____ so it can make a _____ intermediate from its genome.
9. Enveloped viruses are released from a host cell by _____. In this process, the host cell _____ (dies/remains alive).
10. Cells that divide uncontrollably are said to be _____; a mass of such cells forms a _____. If these cells are malignant, they spread in a process called _____ to cause _____.
11. The two types of cell cultures are _____ and _____.

Short-Answer Questions for Thought and Review

1. List the steps in lytic phage replication. For each step, explain in one sentence what happens to the virus.

2. Summarize the key differences between a lytic replication cycle and a lysogenic replication cycle.

3. List the differences that exist between phage replication and the replication cycle of an enveloped animal virus.

4. State the two major differences between latency and lysogeny.

Critical Thinking

1. Why would a dichotomous key be useless for helping a laboratory technician to identify viruses in a patient sample?
2. Assume an individual has the unfortunate luck of being infected with both a bacterial pathogen and a virus at the same time. Assuming the same number of each agent was introduced and that both can actually establish infection in the human host, which microbe will be the most likely cause of disease? Why?
3. If an enveloped virus loses its envelope before it can get to a new host cell, will it be able to infect that cell? Answer yes or no and explain.
4. For the genome types listed in Table 13.3, rank them in terms of which viruses would have the least complications or problems in replicating inside a host cell. The first virus on your list should have the easiest time replicating, and the last virus on your list should have the hardest.
5. There is some debate as to whether or not viruses are alive. Viroids and prions, however, leave little doubt as to the fact that they are not alive. Why not? (Hint: Look at the comparisons outlined in Table 13.5.)

Concept Building Questions

1. Based on the concepts of protein structure, function and chemical bonding discussed in previous chapters, explain how some viruses, such as HIV, have very specific host cell requirements whereas other viruses, such as rabies, can infect many cell types. How is this specificity/non-specificity achieved?
2. Based on our studies of microbial genetics, explain why it is nearly impossible to detect the presence of a lysogenic phage embedded in the host genome. Can these infections ever be "cured"? What about latent animal virus infections?