Taxonomy

3

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This chapter addresses the classification and taxonomy of viruses with special attention to viruses that show pneumotropic properties. Information provided in this chapter supplements that provided in other chapters in Parts II–V of this volume that discuss individual viral pathogens.

3.1 Brief Introduction

Taxonomy may be defined as a logical discipline for the identification and classification of biological entities based on objective, measurable characteristics of relevant entities. Useful taxonomic systems should be broadly applicable across diverse types of biological groups. They should also be flexible, so that new data from technological advances may be integrated into the classification scheme. Primary goals of systemic taxonomy, regardless of biological discipline, include the following:

- Establishing groups (taxa) that reflect varying degrees of evolutionary relatedness among the different biological entities studied
- Establishing criteria for assignment of known or unknown clinical isolates to a given group
- Establishing a clear and unequivocal nomenclature

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The origins of biological taxonomy are firmly rooted in botany and zoology. Early taxonomic systems relied on gross characteristics, like biological niche, internal and external morphology, reproductive strategies and compatibilities, and fossil records. The seminal works of the Swedish botanist Carl Linnaeus used a hierarchical scheme to represent biological relatedness and established the simplified binomial system of nomenclature that serves as the basis for modern classification systems. The modern scientific classification in biology is designed to describe all biological entities within a hierarchy consisting of the following taxa:

Domain → Kingdom → Phylum → Class → Order → Family → Genus → Species

A basic assumption for the establishment of such a hierarchy assumes that all biological entities have evolved from a single common cellular life-form. Different biological entities have evolved as a result of accumulated changes in DNA that have provided survival advantages in different ecological niches. Species may be classified on the basis of phylogenetic and evolutionary relatedness: members of a given species are the most closely related, different species within a single genus are more closely related to each other than to a species within a different genus, and so on. Newer technologies like microscopy, improved biochemical and physiological analysis, and advanced protein and molecular analytical methods have resulted in an enormous

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expansion of characteristics that may be studied for the classification of biological entities and validation of taxonomic systems (Woese et al. 1990).

3.2 The Classification and Taxonomy of Viruses

There are a number of excellent texts that discuss the clinical and laboratory aspects of virus biology (Knipe and Howley 2007; Richman et al. 2009; Versalovic 2012). Though viruses are certainly "biological entities," they are fundamentally different from the cellular life-forms classified by previous taxonomic schemes. Viruses have no autonomous metabolic or replicative ability; they are completely dependent on cellular life-forms. However, within their biological milieu, viruses do replicate and evolve, and they are composed of the same types of organic macromolecules as are cellular life-forms. Because of their intimate relationship with cellular life-forms, it seems legitimate to integrate the schemes for classification of viruses with the schemes used for biological classification of cellular life-forms (Lefkowitz 2012). Initially, various features, like host range, cross-immunity, clinical disease, and pathologic features, were used to classify viruses. Technological advances have led to more detailed and integrated classification, taxonomy, and phylogenetic characterization (evolutionary relatedness) of viruses. Sophisticated nucleic acid sequence analysis has emerged as a powerful tool for virus classification and phylogenetic determination, in spite of some limitations (Holmes 2008; McCormack and Clewley 2002; Zanotto et al. 1996).

A robust system for classification of viruses developed by David Baltimore has gained wide acceptance (Baltimore 1971). Classification is based on the genomic nucleic acid used by the virus (DNA or RNA), strandedness (single or double stranded), and method of replication. The system has been used to define seven classes of viruses:

Class I: Double-stranded DNA (dsDNA)
Class II: Single-stranded DNA (ssDNA)
Class III: Double-stranded RNA (dsRNA)
Class IV: Single-stranded RNA (ssRNA), positive-sense
Class V: Single-stranded RNA, negative-sense
Class VI: Positive-sense ssRNA that replicate by reverse transcription through a DNA intermediate
Class VII: dsDNA viruses that replicate by reverse transcription through an ssRNA intermediate

The International Committee on Taxonomy of Viruses (ICTV) was established by the International Union of Microbiological Societies to oversee and communicate critical issues related to the classification of viruses. This committee was charged with the following:

- Developing a taxonomy for viruses (Eberhard 2004; van Regenmortel and Mahy 2004)
- Developing a nomenclature for specific viral taxa, including virus species
- Communicating taxonomic decisions through periodic summary reports, meetings, and journal publications, e.g., Virology Division News in the Archives of Virology
- Establishing and maintaining an index of virus names (International Committee on Taxonomy of Viruses 2012)

There are six subcommittees of the ICTV through which proposals related to taxonomy are submitted. These experts evaluate evidence related to proposals for taxonomic changes and make recommendations for final approval by vote of entire ICTV membership. The ICTV recognizes only five of the taxa of classic biological taxonomy:

Order (suffix : -*virales*) → **Family** (suffix : -*viridae*) → **Subfamily** (suffix : -*virinae*) → **Genus** (suffix : -*virus*) → **Species**

The primary classification of viruses is into species. A virus species is defined as a polythetic class of viruses that constitute a replicating lineage and occupy a specific ecological niche (International Committee on Taxonomy of Viruses 2002). In polythetic classifications, group members share a number of characteristics, but no single characteristic is necessary

or sufficient to define members of the group. Higher-level taxa are monothetic, i.e., there are characteristics that are necessary and sufficient to define members of the class. It is important to note that not all viruses can be assigned through all taxonomic levels. Virus species may be assigned to a genus or remain unassigned. Similarly, a genus may be assigned to a family or subfamily, or remain unassigned, and so on up the taxonomic hierarchy. Each genus has a *type species*. The type species is the virus that necessitated the creation of the genus; it is always linked to the genus.

In the most recent publication (2012), the ICTV recognized 7 orders, 96 families, 22 subfamilies, 420 genera, and 2,618 species. Important characteristics used by the ICTV to define and classify viruses within these taxa include the following:

- *Susceptible Host Range*: Most viruses have a restricted range of hosts which they are able to infect.
- *Virus Structure*: The viral genome is surrounded by a protective shell of proteins called a capsid. The capsid may also enclose proteins, like reverse transcriptase or proteins required for organization of the nucleocapsid. A nucleocapsid refers to a viral nucleus surrounded by an intact capsid. The nucleocapsids of certain viruses are also surrounded by an envelope of host-derived membranes. The complete virus particle is referred to as a virion.

  Icosahedral capsids are very common; these quasi-spherical shells are composed of 20 identical equilateral triangles with 30 edges and 12 vertices. Icosahedral capsids are very efficient geometrically (internal volume versus protein content) and genetically (many small sides require fewer and smaller genes to code for capsid proteins). The nucleocapsid proteins of some viruses, like the influenza viruses, form helical tubes with the nucleic acid incorporated directly into the helical structure.

  The nucleocapsids of some viruses are surrounded by envelopes composed of lipid bilayers and host- or viral-encoded proteins. Envelopes are typically acquired by budding of the nucleocapsid through a virally modified portion of a specific host-cell membrane (plasma, endoplasmic reticulum, Golgi, nucleus).

  The shape of the virus nucleocapsid or intact virion is usually determined by electron microscopy. The shape and dimensions of the nucleocapsid and intact virion, and the presence or absence of an envelope, are useful characteristics for classifying viruses.
- *Genome*: The viral genome is either DNA or RNA; the nucleic acids may be single or double stranded. The genome size may be expressed in terms of kilobases (kb) for single-stranded genomes or kilobase pairs (kbp) for double-stranded genomes. The sequence of genes of positive-sense ssRNA may be directly translated by the host into viral proteins. The sequence of negative-sense ssRNA is complementary to the coding sequence for translation, so mRNA must be synthesized by RNA polymerase, typically carried within the virion, before translation into viral proteins. The sequence of positive-sense ssDNA is the same as that of the mRNA coding for viral proteins; negative-sense ssDNA is complementary to mRNA and may be transcribed into mRNA for viral protein synthesis. Ambisense single-stranded nucleic acids use both positive-sense and negative-sense sequences. The viral nucleic acid may be linear or circular; the nucleic acid may be in the form of a single molecule or broken into two or more segments.

  In addition to the type of nucleic acid, the size of the viral genome, measured in number of bases or base pairs, is an important characteristic used for classification.
- *Nucleic Acid Sequence Analysis*: The analysis of specific viral nucleic acid sequences is increasingly used as a powerful tool for taxonomic assignment and assessment of evolutionary relatedness. The utility is greatest for related groups of viruses (Lauber and Gorbalenya 2012a, b), but has been challenging for more divergent groups of viruses. Sequence analysis alone has not provided a

reliable single criterion on which all viruses may be classified. Construction of a universal phylogenetic tree for viruses, as has been proposed for cellular life-forms, may not be possible for viruses. It is not clear that all viruses emerged from a single progenitor virus; there is evidence for multiple, independent origins of existing viruses. Phylogenetic analysis using nucleic acid sequences is further complicated by recombination, reassortment, incorporation of host nucleic acid sequences, and other factors (Domingo 2007; Holmes 2011).

Currently, expert consensus, considering laboratory, phenotypic, clinical, and other characteristics, remains the most accurate and robust method for the classification and taxonomic assignment of viruses. Note that the formal names assigned at all taxonomic levels are italicized, while the common names, which are often used clinically, are not italicized. The viruses that have been associated with human infections are shown in Table 3.1.

3.3 Viruses and the Lung

Among the families of viruses able to infect humans and other vertebrate hosts, there are many species that target and cause disease in the lung. These viruses commonly use airborne transmission as an effective mode of transmission between an infected host and a new susceptible host. Characteristics of viruses that directly or indirectly cause pulmonary disease are discussed in this section.

Adenoviridae: Adenoviruses are pathogenic for humans and other vertebrate species. A structural protein at each of the 12 of the icosahedral nucleocapsid vertices anchors a rodlike projection with a terminal knob, which interacts with specific host surface receptor molecules and which confers the hemagglutination pattern and tissue tropism for the different groups of adenoviruses. The genome encodes ~40 genes (Davison et al. 2003a), including common genes and species-specific genes. Genes are grouped into early, delayed early, and late transcribed genes. The genome contains inverted repeat sequences at both ends. Sequences of both DNA strands are transcribed to mRNA; mRNA splicing is used for expression of many adenovirus genes.

The family *Adenoviridae* has not been assigned to an order. Within this family, there are five genera. The seven species that cause human infection are *Human adenovirus A, B, C, D, E, F,* and *G*, all within the *Mastadenovirus* genus; there are 57 accepted serotypes (Buckwalter et al. 2012). Endemic respiratory infections are most commonly caused by serotypes of *Human adenovirus C* (the type species of the genus); most epidemic respiratory infections are caused by serotypes within species *adenovirus B* and *adenovirus E*.

Arenaviridae: Arenaviruses may cause several hemorrhagic fever syndromes. Specific rodents are the reservoir for each arenavirus; human disease is incidental and is usually transmitted by infectious aerosols. Viruses of this family are enveloped; evenly spaced glycoprotein complexes (a tetramer of viral GP2 with viral GP1 ionically bound as a globular head) are attached to the envelope giving complete virions a studded spherical morphology. Complete virions are ~100 nm in diameter, but show significant pleomorphism (range, 60–300 nm). The genome is divided into two segments which are complexed with nucleoproteins (Peters 2009). Complementary sequences at the 3′ and 5′ ends of each segment result in the formation of two circular nucleocapsids. Arenaviruses use both negative-sense and ambisense coding strategies. Host ribosomes are often incorporated within the envelope of complete virions.

This family of viruses is not assigned to an order. There is one genus, *Arenavirus*, with 25 species that fall into two complexes on the basis of serologic and genetic relatedness. The Old World, or African, species include *Lassa virus* (Lassa fever) and *Lujo virus.* The New World species include *Guanarito virus* (Venezuelan HF), *Junín virus* (Argentine HF), and *Machupo virus* (Bolivian HF). The type species of the genus *Arenavirus* is *lymphocytic choriomeningitis virus*.

Bunyaviridae: Bunyaviruses may cause several hemorrhagic fever syndromes. Viruses

Table 3.1 Virus families associated with human infections

Nucleic acid	Family	Segments	Genome	Nucleocapsid	Envelope
Class I: dsDNA	*Adenoviridae*	Unsegmented	Linear, 30–38 kbp	Icosahedral	No
	Herpesviridae	Unsegmented	Linear, 125–240 kbp	Icosahedral	Yes
	Papillomaviridae	Unsegmented	Linear, 7–8 kbp	Icosahedral	No
	Polyomaviridae	Unsegmented	Circular, 5 kbp	Icosahedral	No
	Poxviridae	Unsegmented	Linear, 130–375 kbp	Ovoid	Yes
Class II: ssDNA	*Anelloviridae*	Unsegmented	Circular, 3–4 kb. Negative-sense	Icosahedral	No
	Parvoviridae	Unsegmented	Linear, 4–6 kb. Ambisense	Icosahedral	No
Class III: dsRNA	*Reoviridae*	10–12 segments	Linear, 19–32 kbp	Icosahedral	No
Class IV: ssRNA, positive-sense	*Astroviridae*	Unsegmented	Linear, 6–7 kb	Icosahedral	No
	Caliciviridae	Unsegmented	Linear, 7–8 kb	Icosahedral	No
	Coronaviridae	Unsegmented	Linear, 27–32 kb	Helical	Yes
	Flaviviridae	Unsegmented	Linear, 10–12 kb	Spherical	Yes
	Hepeviridae	Unsegmented	Linear, 7 kb	Icosahedral	No
	Picornaviridae	Unsegmented	Linear, 7–9 kb	Icosahedral	No
	Togaviridae	Unsegmented	Linear, 10–12 kb	Icosahedral	Yes
Class V: ssRNA, negative-sense	*Arenaviridae*	2 segments	Linear, 11 kb. Ambisense	Sphere	Yes
	Bornaviridae	Unsegmented	Linear, 9 kb	Not defined	Yes
	Bunyaviridae	3 segments	Linear, 11–19 kb. Ambisense	Helical	Yes
	Filoviridae	Unsegmented	Linear, 19 kb	Helical	Yes
	Orthomyxoviridae	6–8 segments	Linear, 10–15 kb	Helical	Yes
	Paramyxoviridae	Unsegmented	Linear, 13–18 kb	Helical	Yes
	Rhabdoviridae	Unsegmented	Linear, 11–15 kb	Bullet shaped	Yes
	Deltavirus	Unsegmented	Circular, 2 kb	Spherical	Yes
Class VI: ssRNA, positive-sense, reverse transcribed	*Retroviridae*	Unsegmented, diploid	Linear, 7–13 kb	Icosahedral (spherical or cone-shaped core)	Yes
Class VII: dsDNA, reverse transcribed	*Hepadnaviridae*	Unsegmented	Circular, partially dsDNA, 3kbp	Icosahedral	Yes

within this family are enveloped with protein complexes (heterodimers of glycoproteins Gn and Gc) anchored to the lipid bilayer. Complete virions are spherical (80–120 nm diameter) with projecting spikes; the pattern of spikes varies among different species. The genome consists of three ssRNA strands, designated short (1–2.2 kb), medium (3.5–6 kb), and long (6.3–12 kb) (Mertz 2009). The RNA is complexed with nucleocapsid protein to form three helical nucleocapsids (small, medium, and large) within the envelope. Some bunyaviruses use negative-sense coding exclusively; some use a combination of negative-sense and ambisense coding.

The family *Bunyaviridae* is not assigned to an order. There are five genera in this family. There are 24 species in the genus *Hantavirus*, including *Andes virus*, *Hantaan virus* (the type species), *Puumala virus*, *Seoul virus,* and *Sin Nombre virus*. Rodents, not arthropods, are the reservoir for species of the genus *Hantavirus*. In the genus *Nairovirus*, there are seven species, including *Crimean-Congo hemorrhagic fever virus* (tick vector). In the genus *Phlebovirus*, there are nine species, including *Rift Valley fever virus* (the type species, mosquito vector) and *Sandfly fever Naples virus* (sandfly vector).

Coronaviridae: Transmembrane proteins produce blunt projections from the surface of coronaviruses, resulting in a "crown-like" appearance on electron microscopic studies (100–160 nm in diameter). Translation of the coronavirus genome is unique and includes production of polyproteins, discontinuous synthesis, overlapping reading frames, ribosomal frame shifting, and post-translational proteolytic processing (Marra et al. 2003; Rota et al. 2003; Theil et al. 2003). The major structural proteins, spike glycoprotein (S), membrane glycoprotein (M), nucleocapsid phosphoprotein (N), hemagglutinin-esterase glycoprotein (HE), and envelope protein (E), are present in all coronaviruses. Nonstructural proteins are encoded in 5–10 unique or overlapping reading frames (Lai et al. 2007).

The human coronaviruses are assigned to the order *Nidovirales*, family *Coronaviridae*, and subfamily *Coronavirinae*. There are four genera and three serological groups. Relevant viruses include *Human coronavirus 229E* and *Human coronavirus NL63* of the genus *Alphacoronavirus* (antigenic group I), *Human coronavirus HKU1, Betacoronavirus 1* and *Severe acute respiratory syndrome-related coronavirus* of the genus *Betacoronavirus* (antigenic group II).

Filoviridae: Filoviruses may cause several hemorrhagic fever syndromes. The filoviruses have a unique threadlike morphology. The helical nucleocapsids are surrounded by an envelope studded by spikes formed by a single type of glycoprotein (GP). The genome consists of a single segment of negative-sense ssRNA that encodes for seven proteins (Kuhn et al. 2010). The presence of gene overlap for several genes is an unusual feature of filoviruses. In ebolaviruses, the surface glycoprotein is encoded by two adjacent reading frames. A truncated version (sGP), which lacks the hydrophobic anchor, results from translation of the upstream reading frame only. This protein is secreted from cells and may serve as a decoy for the host's immunological response. The full-length GP is formed only when the RNA polymerase misreads a poly-U editing site between the reading frames. The full-length GP is inserted, as homotrimers, into the host membranes that will form the virion envelope. A helical nucleocapsid is formed by association of the ssRNA with nucleoproteins. The nucleocapsid is ~50 nm in diameter, with a central axial space ~20 nm in diameter. The nucleocapsid is attached to the envelope by matrix protein. The complete virions are ~80 nm in diameter, but the virion length may vary from 800 to 10,000 nm.

The family *Filoviridae* is assigned to the order *Mononegavirales*. There are two genera within the family *Ebolavirus* and *Marburgvirus*. There are five ebolavirus species, including *Sudan ebolavirus* and *Zaire ebolavirus* (the type species). The genus *Marburgvirus* consists of one species, *Marburg marburgvirus*. Humans and nonhuman primates are susceptible to ebolavirus and marburgvirus infection; the host reservoirs for these viruses are unknown. Humans may be infected sporadically by presumed contact with the host species or by direct contact with virus containing body fluids taken from acutely infected humans or nonhuman primates. Nosocomial and laboratory-acquired infections are well described.

Flaviviridae: Flaviviruses may cause several hemorrhagic fever syndromes. *Hepatitis C virus* is also a flavivirus species. Flaviviruses are surrounded by an envelope studded with dimers of viral E glycoprotein and M protein which give the mature virion a herringbone appearance with icosahedral symmetry. The genome consists of a single segment of positive-sense ssRNA (Chambers et al. 1990; Osatomi and Sumiyoshi 1990). Cyclization of the genome, through hybridization of RNA sequences of the 5′ and 3′ ends of the genome, may be required for mRNA synthesis (Alvarez et al. 2005). There is a long open reading frame that codes for three structural proteins at the 5′ end; downstream of this region are genes for seven nonstructural proteins (Thurner et al. 2004). The positive-sense genome is directly translated into a large polyprotein, which undergoes intra- and post-translational cleavage. Strain evolution and clinical diversity have been driven by a high rate of mutation at replication and through molecular recombination. The nucleocapsid is formed by interaction of genomic RNA with capsid proteins. The complete virion has a spherical morphology approximately 50 nm in diameter.

This family of viruses is not assigned to an order. There are four genera within the family *Flaviviridae*. Within the genus *Flavivirus*, there are 53 species, including *Dengue virus* (Simmons et al. 2012), *Kyasanur Forest disease virus*, *Omsk hemorrhagic fever virus*, and *Yellow fever virus* (the type species). In addition to viruses that cause hemorrhagic fever syndromes, the family *Flaviviridae* includes many species of neurotropic viruses that cause encephalitis and other CNS infections, like *Japanese encephalitis virus*, *St. Louis encephalitis virus*, *Tick-borne encephalitis virus*, and *West Nile virus*. Most human flavivirus infections are transmitted by mosquitoes or ticks.

Hepatitis C virus (HCV) is the type species of the genus *Hepacivirus* in the family *Flaviviridae*. The physical properties of HCV have not been as well defined as other flaviviruses because there is no efficient method for in vitro replication of HCV. Virion morphology is consistent with other flaviviruses; complete, enveloped virions have a diameter of 55–65 nm. The single segment positive-sense ssRNA is ~9.6 kb in length (Hijikata et al. 1991). A single open reading frame is flanked by highly conserved regions at the 5′ and 3′ ends. Cap-independent protein synthesis, typical of *Flavivirus* species, is initiated at an internal ribosomal entry site (IRES) within the 5′ untranslated region. This results in synthesis of a polyprotein that undergoes cleavage and further processing during and after translation. A unique and highly conserved sequence upstream of the IRES interacts with liver-specific microRNA and is required for efficient replication. Circulating HCV is associated with host LDL/VLDL, which may play a role in delivery of virions to hepatocytes.

The error-prone RNA polymerase and high replication rate of HCV has resulted in a great genetic diversity and heterogeneity of clinical isolates. HCV isolates can be grouped by genotypic analysis into six groups and many subgroups. There are differences with respect to responses to antiviral therapy among the genotypes, but intrinsic virulence is similar. The vast majority of strains in the United States are genotypes 1a, 1b, and 2, whereas Central African strains are almost exclusively genotype 4.

Hemorrhagic Fever (HF) Syndromes: Viral hemorrhagic fever syndromes may be caused by many species of viruses from four different families: *Arenaviridae, Bunyaviridae, Flaviviridae* and *Filoviridae*; all are single-stranded RNA viruses. See the discussions above for specific information related to these virus families.

Typical symptoms of viral hemorrhagic fever infection include fever, malaise, hypotension, and coagulation defects. With the exception of dengue, the other HF viral agents are maintained in nonhuman vertebrate hosts; humans are coincidental, dead-end hosts. In dengue, human infection is maintained through a mosquito vector. The epidemiologic distribution of disease reflects the geographic range of the reservoir host.

HF viruses primarily infect dendritic cells, macrocytes, and monocytes, which are present in virtually all tissues and organ systems; parenchymal cells may also be susceptible to infection,

depending on the virus. Infected cells release mediators that result in marked increased vascular permeability, compromising the function of critical organ systems. Suppression of cellular type 1 interferon response is a significant contributor to pathogenesis (Habjan et al. 2008).

Hepadnaviridae: In the family *Hepadnaviridae*, there are two genera, *Avihepadnavirus* (two species) and *Orthohepadnavirus* (four species); *hepatitis B virus* (HBV), the type species of *Orthohepadnavirus*, is only human pathogen in family. The family *Hepadnaviridae* is not assigned to an order. Eight distinct HBV genotypes (A–H) and subtypes can be recognized on the basis of antigenic or sequence variation. The genotypes show geographic and ethnic variability; the HBV genotype influences the severity and outcome of disease (Garfein et al. 2004; Lin and Kao 2008).

The complete, enveloped HBV virion (Dane particle) is 42–47 nm in diameter. The icosahedral nucleocapsid (~28 nm in diameter) of the virion contains a single molecule of partially double-stranded DNA with a DNA-dependent polymerase covalently linked to the 5′ end of the complete DNA strand, hepatitis B e antigen (HBeAg) and hepatitis B core antigen (HBcAg). The nucleocapsid is surrounded by an envelope derived from host-cell membrane and viral envelope proteins, including hepatitis B surface antigen. The genome of HBV is a circular, partially double-stranded DNA molecule which is replicated by a unique process of reverse transcription of an RNA intermediate. The minus DNA strand runs the entire length of the HBV genome; the plus strand covers only about two-thirds of the genome. The genome is replicated by synthesis of a full-length ssRNA transcript (pre-genomic RNA), followed by dsDNA synthesis by reverse transcription of the ssRNA by viral-encoded reverse transcriptase/DNA polymerase. All viral proteins are also transcribed from the minus DNA strand. There are four overlapping open reading frames, all read in the same direction (Liang 2009).

Herpesviridae: The herpesvirus species associated with human infections (HSV-1, HSV-2, CMV, EBV, VZV, HHV-6, HHV-7, and HHV-8) belong to the family *Herpesviridae* within the order *Herpesvirales*. There are four subfamilies of the *Herpesviridae*: *Alphaherpesvirinae* (5 genera), *Betaherpesvirinae* (4 genera), *Gammaherpesvirinae* (4 genera), and a single genus in an unassigned subfamily. Specific human herpesviruses are discussed in the sections below.

The herpesviruses are double-stranded DNA viruses. The icosahedral capsid (~100 nm diameter) is surrounded by an envelope studded by a variety of short glycoproteins. The nucleocapsid is a dense toroid complex with an outer diameter ~70 nm and inner diameter ~18 nm. An irregular "tegument" fills the space between the envelope and capsid. Depending of the thickness of the tegument layer, complete virions range in size from ~125 to >250 nm. The size and organization of the dsDNA genome varies among the species causing human disease (McGeoch et al. 2006). The genomes of human herpesviruses include unique sequences and repeated sequences. Though the genomes are linear in virions, they circularize in the nucleus of infected cells, which is mediated through repeat sequences at both ends of the dsDNA genome. For HHV6 and HHV7 (class A genome), a large unique sequence region is flanked by a region that is repeated at both ends of the linear strand of dsDNA. The genome of EBV and the Kaposi's sarcoma-associated herpesvirus (class C genome) have smaller left and right terminal repeat sequences, while repeat sequences R1 to R4 divide the unique sequence nucleic acid into four discrete regions. For VZV (class D genome), a large terminal sequence is inverted and inserted into the genome, resulting in a large unique sequence region (UL) and a small unique sequence region (US). HSV-1, HSV-2, and CMV (class E genomes) are the most complex. There are repeat sequence regions at both ends of the linear dsDNA molecule. The unique sequence dsDNA is divided into UL and US regions by a sequence composed of juxtaposed copies of the terminal repeat sequences inserted in an inverted orientation.

Typical of dsDNA viruses, a large number of proteins are produced by various herpesviruses. The organization of the coding regions is complex, with 3′ and 5′ reading frames, gene overlap, spliced genes, and intron regions. Forty genes are conserved among the α-, β-, and γ-herpesviruses. These core genes are divided among seven gene blocks (Albà et al. 2001); within each block the order and polarity of genes are conserved, including genes for gene regulation, nucleotide metabolism, DNA replication, virion maturation, envelope glycoprotein synthesis, and capsid, fusion and tegument protein synthesis.

Diseases caused by human herpesviruses range from systemic to localized infection of virtually all organ systems, although the host-cell range and typical disease characteristics vary by species. A characteristic of herpesvirus infections is latency, which is commonly associated with reactivation and symptomatic infections (e.g., shingles). While active infection with herpesviruses results in the destruction of the infected host cell, latently infected cells remain viable. In latently infected cells, the viral genome forms circularized molecules within the host nucleus with limited expression of viral genes.

- *Cytomegalovirus*: Human cytomegalovirus (hCMV) is the most complex human herpesvirus. The complete virions of human cytomegalovirus range in diameter from ~200 to 300 nm. The Golgi-derived envelope is studded with 20 or more virally encoded glycoproteins. The icosahedral nucleocapsid (~125 nm) includes five capsid proteins enclosing a class E genome (~230 kbp linear dsDNA) (Davison et al. 2003b; Dunn et al. 2003), as described above. The hCMV tegument is composed of at least 27 virally encoded proteins and other viral and host-cell macromolecules.

  The species designation of hCMV is *Human herpesvirus 5* in the genus *Cytomegalovirus* (for which it is the type species) and subfamily *Betaherpesvirinae*.
- *Epstein-Barr Virus*: The morphology of Epstein-Barr viruses (EBV) is typical of other herpesviruses: a single glycoprotein is predominant in the envelope. The EBV virions contain a type C genome (172 kbp). The genome encodes more than 70 proteins, including the core herpesvirus genes, as well as species-specific genes (Baer et al. 1984). The primary target of EBV is the B-lymphocyte, in which long-term latency is established. Lytic infection of epithelial cells is primarily responsible for transmission of infection.

  The species designation of EBV is *Human herpesvirus 4* in the genus *Lymphocryptovirus* (for which it is the type species) and subfamily *Gammaherpesvirinae*. Oncogenic potential is a characteristic of the *Gammaherpesvirinae* and is well documented for EBV. There are two types of EBV, which can be detected by serological reactions directed against nuclear antigens. Type 1 EBV isolates are most prevalent in the United States and other developed countries; both type 1 and 2 EBVs are seen in African isolates.
- *Herpes Simplex Viruses*: Complete HSV virions are ~225 nm in diameter with a spiked outer surface caused by 11 viral glycoproteins. The icosahedral nucleocapsid is eccentrically placed within the envelope. The HSV linear genome (150–155 kbp) has a class E organization (Mocarski and Roizman 1981). Reading frames are present on both DNA strands. Individual reading frames may be embedded in larger reading frames, and antisense reading frames are used for several proteins. Several transcripts are formed by RNA splicing. Transcription switches from host cell to viral genes within several hours after infection.

  Two distinct herpes simplex viruses can be recognized by antigenic and genetic differences (Dolan et al. 1998): *Human herpesvirus 1* and *Human herpesvirus 2*. These viruses are assigned to the subfamily *Alphaherpesvirinae* and the genus *Simplex virus*; *Human herpesvirus 1* is the type species. Human herpes simplex viruses are able to infect cells of many different organ systems.
- *HHV-6 and HHV-7*: The genome (~170 kbp for HHV-6; ~150 kbp for HHV-7) is contained by a 90–110 nm icosahedral capsid.

Tegumented capsids released from the nucleus are enveloped by cytoplasmic vacuoles; six viral glycoproteins are embedded in the envelope of complete virions. Cell-free virions are 170–200 nm in diameter. The class A genome (Gompels et al. 1995) of these viruses has the simplest organization and lowest %G+C content compared to the other herpesviruses. Reading frames are present on each strand of the dsDNA. Core herpesvirus proteins are clustered near the center of the strands, while species-specific genes are located toward the ends of the strands (Braun et al. 1997).

HHV-6 and HHV-7 are assigned to the genus *Roseolovirus* in the subfamily *Betaherpesvirinae*, family *Herpesviridae*, and order *Herpesvirales*. There are two distinct HHV-6 species: *Human herpesvirus 6A* (the Roseolovirus type species) and *Human herpesvirus 6B*. HHV-6B is the agent of exanthem subitum. There is a single HHV-7 species, *Human herpesvirus 7*, which is also a cause of exanthem subitum. T-lymphocytes are the primary target cell of HHV-6 and HHV-7 viruses.

- *Kaposi's Sarcoma-Associated Herpesvirus (HHV-8)*: The complete virions of Kaposi's sarcoma-associated herpesvirus (KSHV) have a diameter ~100 nm. In addition to virus-specific proteins, the tegument also carries viral mRNAs, probably the result of passive incorporation during the cytoplasmic envelopment process (Bechtel et al. 2005). The envelopes of complete virions bear KSHV-specific glycoproteins. The genome (~170 kbp) has class C organization (Russo et al. 1996) typical of *Gammaherpesvirinae*. The conserved herpesvirus genes are clustered in four blocks; KSHV-specific genes are typically distributed in the regions outside and between these blocks (Renne et al. 1996).

  The KSHV species designation is *Human herpesvirus 8*, which is assigned to the genus *Rhadinovirus* within the subfamily *Gammaherpesvirinae*. The virus has tropism for B-lymphocytes and is implicated in all forms of Kaposi's sarcoma. Four clades, A–D, with distinctive geographical distributions, have been identified by genotypic analysis; the A and C clades cluster together and are most typical for isolates from Europe and the United States.
- *Varicella-Zoster Virus (VZV)*: The dense core of VZV is enclosed in an icosahedral capsid (80–120 nm diameter), which is surrounded by an amorphous tegument. The envelope may be derived from multiple types of host-cell membranes during transit from the nucleus through the cytoplasm; specific viral-encoded glycoproteins are embedded in the envelope of the complete virions, which may be spherical or pleomorphic (180–200 nm in diameter). VZV has a class D dsDNA genome (~125 kbp) (Clarke et al. 1995; Davison 1984), resulting in production of two isomeric genomic forms by infected cells through inversion of the US region (Ecker and Hyman 1982). The genome encodes more than 70 proteins. The organization includes grouping of several genes into single transcription units, genes with overlapping reading frames, and spliced segments (Davison and Scott 1986).

  The species designation for VZV is *Human herpesvirus 3*. It is the type species of the genus *Varicellovirus* within the subfamily *Alphaherpesvirinae*. There is only a single serotype of VZV. For epidemiologic purposes, VZV isolates may be genotyped on the basis of minor differences in DNA sequence; different genotypes may be classified as European, Japanese, or Mosaic (Loparev et al. 2004). The host range of VZV is restricted to cells of humans or other primates; in humans, VZV has tropism for human T-lymphocytes and establishes latent infection in the cells of the dorsal root ganglia.

Orthomyxoviridae: Influenza viruses belong to the family *Orthomyxoviridae*. They are polymorphic; viruses may be spherical (~100 nm diameter) or filamentous. Complete virions are surrounded by an envelope derived from the host cytoplasmic membrane. Viral hemagglutinin and neuraminidase proteins are embedded in the envelope resulting in characteristic 10–14 nm spikes projecting from the surface of virions. In addition to the HA and NA protein, M2 protein is embedded into the envelope of *Influenza*

A viruses; NB and BM2 proteins are embedded into the envelopes of *Influenza B* viruses. The matrix protein (M1) is located just below the envelope. The nucleocapsid is composed of viral RNA and nonstructural proteins, including ribonucleoproteins and polymerases.

The genome of influenza viruses is composed of negative-sense ssRNA. All viral RNA synthesis occurs in the nucleus of the host cell. The A and B influenza virus genomes are composed of eight segments, while the *Influenza C* virus genome consists of seven segments (Hayden and Palese 2009). The segments range in size from ~900 to 2,300 nucleotides in length. Each segment codes for one or more viral proteins (McCauley et al. 1983). The 3′ and 5′ ends of each segment contain noncoding, regulatory regions (Fujii et al. 2005). The three largest segments code for various components of RNA polymerase; the PB1 segment of *Influenza A* virus has a second open reading frame that encodes the pro-apoptotic protein PB1-F2. In influenza types A and B, the fourth and sixth segments encode for the surface hemagglutinin (HA) and neuraminidase (NA) glycoproteins, respectively. The *Influenza A* surface protein M2 is encoded by the seventh segment; Influenza B surface protein NB is encoded by the sixth segment, while the BM2 is encoded by the seventh segment. The fifth segment of both A and B influenza viruses encodes for the RNA-binding nucleoprotein (NP). The matrix protein M1 is encoded by the seventh segment of both viruses. The eighth and smallest RNA segment of influenza A and B viruses encodes for NS1, a multifunctional protein with interferon antagonistic properties and NEP/NS2 protein which is involved in transport of vRNPs across the nuclear membrane of the host cell.

The *Orthomyxoviridae* have not been assigned to an order. There are six genera: *Influenza A*, *Influenza B*, *Influenza C*, *Isavirus, Quaranjavirus,* and *Thogotovirus*. The influenza virus genera each contain a single species: *Influenza A virus*, *Influenza B virus,* and *Influenza C virus*. Most human infections are caused by influenza A and influenza B viruses.

Names of clinical isolates of human influenza isolates include the species of origin, isolation location, number of the isolate, and year of isolation; *Influenza A* virus isolates also include the hemagglutinin (H1 to H16) and neuraminidase (N1 to N9) subtypes (Atmar and Lindstrom 2012). For example, A/California/7/2009 (H1N1), A/Victoria/361/2011 (H3N2), and B/Wisconsin/1/2010 viruses were recommended for the 2012–2013 seasonal influenza vaccine. Large outbreaks have only occurred with H1, H2, and H3 and neuraminidases N1 and N2 viral subtypes. Antigenic drift and antigenic shift contribute to reinfection with influenza viruses (Taubenberger and Kash 2010). Antigenic drift is caused by a gradual accumulation of point mutations in hemagglutinin and neuraminidase genes, which result in minor antigenic changes in these proteins. Antigenic shift is caused by a virus created by reassortment of influenza virus RNA segments during coinfection of a host, usually with a human influenza virus and an avian or swine influenza virus or through introduction of a non-human influenza virus strain into human populations after mutation during a host-species infection creates a new isolate permissive for interspecies transmission.

Papillomaviridae: The papillomaviruses (PVs) represent a large (and growing) family of viruses that currently includes 30 different genera and 69 species; the taxonomy has undergone significant reorganization in recent years (Bravo et al. 2010). The oncogenic potential of human papillomaviruses is well established. PVs are non-enveloped; virions are icosahedral with diameters of 50–55 nm. The capsid contains two structural proteins, L1, the most abundant viral protein, and L2. The PV genome consists of a single molecule of circularized dsDNA (Zheng and Baker 2006). The open reading frames for all viral genes are located on only one of the DNA strands, and transcription proceeds in a single direction. There are eight early (E) open reading frames that encode for regulatory proteins that control viral metabolism and DNA synthesis. The E6 proteins of high-risk HPV types have anti-apoptotic effects and interfere with p53 regulatory function in infected host cells (Howley et al. 1990). Two late (L) reading

frames encode for synthesis of the structural proteins L1 and L2.

Epithelial cells of a wide variety of vertebrate hosts are susceptible to papillomavirus infection, but the different host species are only susceptible to species-specific viruses. Papillomaviruses have been classified according to susceptible host species and the type of disease produced, but comparison of sequence differences of the L1 reading frame has provided a more detailed description of papillomavirus phylogeny (de Villiers et al. 2004). The family *Papillomaviridae* is not assigned to an order. Human pathogens are clustered within five papillomavirus genera.

Paramyxoviridae: The Paramyxoviruses are enveloped (host cytoplasmic membrane) with an unsegmented negative-sense ssRNA genome (15–19 kb). The viral RNA serves as template for synthesis of mRNA and for synthesis of antigenomic (positive-sense) RNA for synthesis of new viral negative-sense RNA for new virions. There are six to ten genes; genes for the six major proteins are linked in the following 3′ to 5′ order: nucleocapsid (N) → phosphoprotein (P) → matrix (M) → fusion (F) → hemagglutinin/neuraminidase (HN) → large polymerase (L). There is an untranslated leader sequence at the 3′ end and untranslated trailer sequence at the 5′ end. The genes are separated by untranslated sequences and do not overlap, with the exception of the M and L genes of *Human metapneumovirus*. Translation is initiated at the 3′ end and proceeds directly through to the 5′ end. Because the RNA polymerase is unstable and may detach at the untranslated regions between genes, there is a gradient in the concentration of gene products from 3′ to 5′. In different species, other proteins are produced by additional small genes, mRNA editing, or overlapping reading frames within the P gene. The V and C proteins regulate viral RNA transcription and also interfere with host interferon signaling and other aspects of the immune response to paramyxovirus infection (Andrejeva et al. 2004; Durbin et al. 1999; Swedan et al. 2009).

Formation of the nucleocapsid core is constrained by a required association of one N protein to every six genomic nucleotides (Kolakofsky et al. 2005; Skiadopoulos et al. 2003). The resulting helical structure has a diameter of 18 nm with a 4 nm central core. P proteins (a polymerase cofactor) are attached to this rigid rod and serve as attachment of L proteins, which interact to provide enzymatic activity for RNA synthesis. This core structure, rather than free genomic RNA, serves as the template for mRNA and antigenomic RNA synthesis. The paramyxovirus M proteins surround and organize the nucleocapsid and interact with the cytoplasmic tails of transmembrane envelope proteins. The envelope formed from modified host-cell plasma membranes is studded by viral protein complexes, including HN proteins, which mediate virion attachment to target cells, and F protein, which mediates pH-independent fusion of the viral envelope and cell cytoplasmic membrane.

The *Paramyxoviridae* are one of the four families within the order *Mononegavirales* and include significant and frequent pathogens of humans and animals. There are two subfamilies: the *Paramyxovirinae* and the *Pneumovirinae*. There are seven genera and thirty-one species in the subfamily *Paramyxovirinae* and two genera and five species in the *Pneumovirinae* subfamily.

- *Henipahviruses:* In the subfamily *Paramyxovirinae*, there are two species within the genus *Henipahvirus*: *Hendra virus* (HeV) and *Nipah virus* (NiV); HeV is the type species. Henipahvirus virions are pleomorphic (spherical to helical forms). Electron microscopy of *Hendra virus* shows a "double-fringe" appearance due to short and long surface projections (Hyatt et al. 2001). Complete virions range in size from 40 to 1,900 nm in longest dimension. The genome (~18 kb) includes genes typical of *Paramyxoviridae*. Long untranslated sequences are attached to the 3′ end of five of the six genes, resulting in the larger genome size of Henipahviruses compared to other Paramyxoviruses (Eaton et al. 2007; Wang et al. 2000). The P gene also codes for V and W proteins by mRNA editing and C protein by a shifted reading frame.

  Henipahviruses are assigned to the family *Paramyxoviridae* and subfamily

Paramyxovirinae. Henipahvirus infections are zoonotic; fruit bats are the presumed reservoir for *Hendra virus* infections, while fruit bats (Johara *et* al. 2001) or domesticated pigs are the presumed reservoir for *Nipah virus* infections.

- *Human Metapneumovirus:* Three transmembrane glycoproteins, G, F, and SH proteins, are embedded in the envelope of *Human metapneumovirus* (hMPV) particles, resulting in 13–17 nm spikes. Complete virions are polymorphic with irregular helical and spherical forms (150–600 nm in diameter). The organization of the hMPV genome (~13.3 kb) is similar to other paramyxoviruses (vdHoogen et al. 2002), with eight nonoverlapping genes: N, P, M, F, M2, SH, G, and L. M2 includes two open reading frames; the function of M2-1 protein is undefined, while M2-2 protein is a regulator of viral transcription. There is no gene for hemagglutinin-neuraminidase; the product of the G gene serves as the major attachment protein.

  Metapneumoviruses are members of the subfamily *Pneumovirinae. Human metapneumovirus* is a species in the genus *Metapneumovirus.* There is a single hMPV serotype, with two antigenic subtypes A and B.
- *Human Respiratory Syncytial Viruses:* The envelope of *Human respiratory syncytial virus* (hRSV) is studded with three viral glycoproteins: G protein (the major attachment protein), F protein, and SH protein (a small hydrophobic protein). Complete virions are pleomorphic (spherical to filamentous forms). The nucleocapsid diameter, 12–15 nm, is smaller than typical for other paramyxoviruses (Hall 2001).

  The hRSV genome is ~15 kb and includes ten genes (Collins and Wertz 1983). The first eight reading frames are nonoverlapping; the last two genes, M2 and L, overlap by 62 nucleotides. In addition to N, P, and L proteins, M2-1 protein, a transcription factor, is associated with the nucleocapsid. The overall organization of the hRSV genome is similar to other paramyxoviruses. In addition to the typical genes, the hRSV genome includes genes NS1 and NS2 (nonstructural proteins that interfere with interferon induction and signaling), SH, and M2-1 and M2-2 (nonstructural proteins involved in regulation of transcription). There is no NH gene; attachment is mediated by the G gene product.

  Human respiratory syncytial virus is the type species of the genus *Pneumovirus* in the subfamily *Pneumovirinae.* There is a single hRSV serotype, *w*ith two antigenic subtypes A and B.
- *Measles Virus:* The envelope of measles viruses contains projections composed of viral hemagglutinin and fusion glycoproteins. Complete virions are pleomorphic and range in size from ~100 to 300 nm. The genome of *Measles virus* is ~16 kb in length (Dowling et al. 1986). C and V proteins are transcribed from the P gene through frame-shifted reading and mRNA editing. There is only a single serotype of *Measles virus*; infection confers lifelong immunity. Formation of intranuclear inclusion bodies and lack of neuraminidase activity are characteristics useful in histologically differentiating *Measles virus* and other morbilliviruses from other species of paramyxoviruses.

  Measles virus is the type species of the genus *Morbillivirus,* within the subfamily *Paramyxovirinae.* In addition to *Measles virus,* there are five other *s*pecies assigned to the genus *Morbillivirus*, including *Rinderpest virus*, a closely related pathogen of cattle.
- *Parainfluenza Viruses:* The lipid envelopes of the human parainfluenza viruses are studded by glycoprotein spikes (hemagglutinin/neuraminidase tetramers and fusion protein trimers). Typical virions are spherical (150–200 nm); filamentous forms may be seen (Henrickson 2003).

  The genome of human parainfluenza viruses is ~15 kb in length with an organization and six reading frames (N, P, M, F, HN, L) typical of the *Paramyxoviridae* (Karron and Collins 2007). There are no overlapping reading frames. Accessory proteins, C (HPIV1 and 3), V (HPIV 2 and 4), and D (HPIV3), are produced by mRNA editing of the P gene.

N proteins are tightly bound to viral and antigenomic RNA; P and L proteins are also bound to the nucleocapsid, forming functional complexes for RNA polymerization and processing.

Human parainfluenza viruses are assigned to two genera in the subfamily *Paramyxovirinae. Human parainfluenza virus 1* and *Human parainfluenza virus 3* are assigned to the genus *Respirovirus;* the *Respirovirus* type species is *Sendai virus. Human parainfluenza virus 2* and *Human parainfluenza virus 4* belong to the genus *Rubulavirus*: the *Rubulavirus* type species is *Mumps virus*. The human parainfluenza viruses are serologically distinct; there is no common antigen among these viruses.

Parvoviridae: Parvoviruses are small, nonenveloped viruses. Complete virions are 18–26 nm in diameter with icosahedral symmetry. Parvoviruses replicate only in dividing host cells or in the presence of a helper virus (e.g., adeno-associated viruses) (Berns 1990). Virions are stable in the environment and thought to transmit infection by attachment to specific receptors of actively dividing cells.

The parvovirus genome is composed of unsegmented ssDNA (Cotmore and Tattersall 1984; Shade et al. 1986; Zhi et al. 2004). Complete virions of different species may contain negative-sense or both negative- and positive-sense DNA in various proportions. There are two major reading frames: one encoding capsid proteins and the other coding for nonstructural proteins. Noncoding sequences at the 3′ and 5′ ends include complementary sequences which result in the formation of hairpin structures that serve to regulate nucleic acid synthesis (Deiss et al. 1990). Various host-cell molecules mediate attachment and infection by parvoviruses. Erythrocyte P antigen is the major receptor for *Human parvovirus B19*. Viruses are taken up by endocytosis, followed by transport into the host-cell nucleus. Viral DNA replication depends on host-cell polymerases during the S phase of host-cell replication.

Human infections are caused by parvovirus B19 and bocavirus (Schildgen et al. 2008; Vicente et al. 2007). The family *Parvoviridae* is not assigned to an order; there are two subfamilies, the *Densovirinae* and the *Parvoviridae*. There are five genera in the subfamily *Parvoviridae*: *Amdovirus* (1 species), *Bocavirus* (2 species including the type species *Bovine parvovirus*), *Dependovirus* (12 species, including adeno-associated viruses), *Erythrovirus* (4 species including the type species *Human parvovirus B19*), and *Parvovirus* (12 species).

Picornaviridae: Infections of the respiratory tract and other organ systems by enteroviruses and parechovirus are well described. Enteroviruses were initially classified on the basis of clinical disease and epidemiology, suckling mouse inoculation, replication in cell culture, electron microscopic studies, physical properties, and the vast range of specific antigenic differences. The major subgroups were poliovirus, coxsackievirus (A and B), and echovirus. A characteristic of these viruses is their relative stability in acidic media and nonionic detergents.

Translation of the positive-sense ssRNA genome is regulated by a 5′ non-translated region (Lindberg and Polacek 2000) that is covalently linked to protein VPg (virion protein, genome linked); the short 3′ noncoding region is polyadenylated. Translation results in synthesis of a single polyprotein, which is cleaved into functional proteins by post-translational processing (Nicklin et al. 1987; Pallansch and Roos 2007). There are three functional regions delimited by ribosomal entry sites. The P1 region codes for capsid proteins, while regions P2 and P3 code for nonstructural proteins. Capsid proteins VP1, VP2, and VP3 are exposed externally and account for the serological diversity of the viruses.

With the advent of molecular phylogenetic analysis, the enteroviruses have been reclassified by the ICTV. Enteroviruses are in the order *Picornavirales*, family *Picornaviridae,* and genus *Enterovirus*. The enteroviruses have been assigned to 12 species, including *Human enterovirus A* (17 serotypes including coxsackieviruses and enteroviruses), *Human enterovirus B* (56 serotypes, including coxsackieviruses, echoviruses, and enteroviruses), *Human enterovirus C* (the type species; 16 serotypes including coxsackieviruses, all human polioviruses, and enteroviruses), and *Human enterovirus D* (3 enterovirus serotypes). In addition to the enteroviruses, the

genus *Enterovirus* also includes 3 rhinoviruses species, *Human rhinovirus A*, *B,* and *C*, and more than 100 serotypes.

Also within the family *Picornaviridae* is the genus *Parechovirus*. *Human parechovirus* is the type species for the genus. There are 14 parechovirus serotypes.

Polyomaviridae: Polyomaviruses may infect a variety of primate and non-primate vertebrate host species; the oncogenic potential of polyomaviruses is well established (White and Khalili 2004). Sialic acid and/or gangliosides on the host-cell membranes serve as receptors for attachment of human polyomavirus. Though these molecules are widespread on human cells, there is a restricted tropism. Respiratory epithelial cells and cells of lymphoid origin are the likely targets for initial infection, followed by hematogenous spread to target organs. The virions are non-enveloped; the icosahedral capsids (40–45 nm diameter) are composed of three proteins (VP1, VP2, and VP3), which enclose the circular dsDNA genome (~5 kbp). The genome is divided into three regions. The early region encodes for proteins involved in viral processes that occur prior to DNA replication, including T (tumor) antigens (Benjamin 2001). The late region encodes for proteins involved in processes that primarily occur after DNA replication. The early and late regions do not overlap and are transcribed from opposite strands of the viral DNA and in opposite directions. A number of viral proteins are encoded as a result of alternative splicing and other posttranslational modifications of mRNA.

Polyomaviruses are members of the family *Polyomaviridae*, which is not assigned to an order. There is 1 genus, *Polyomavirus*, and 13 species, including the human pathogens *BK polyomavirus* and *JC polyomavirus* and *Simian virus 40* (type species).

Retroviridae: The retroviruses are a unique group of viruses, including Human Immunodeficiency Virus types 1 and 2 and Human T-cell Leukemia Virus type 1; they may infect a wide range of vertebral host species. The human immunodeficiency viruses and human T-cell leukemia virus 1 are able to cause disease in humans. These RNA viruses use a unique replication cycle that uses a "reverse flow" of genetic information from RNA to DNA: viral RNA is reverse transcribed and converted into a dsDNA copy of the viral genome which is integrated into the host-cell genome. Integration of the proviral DNA allows the viruses to establish persistent, presumably lifelong, infection. Another consequence of insertion of the viral DNA is functional mutation of the host genome at the site of insertion which may alter the host gene or regulation of a gene's expression; the oncogenic potential of retroviral infection is well described in humans and other vertebrate host species.

The electron microscopic morphology of retroviruses shows a dense nucleocapsid core (cylindrical or cone shaped) (Chrystie and Almeida 1988; Gelderblom et al. 1989). Viruses are functionally diploid: the core includes two copies of the positive-sense ssRNA genome, which are closely complexed with viral nucleoproteins. The sequences of the two ssRNA molecules may differ because of errors in transcription of new genomic ssRNA molecules during replication. The core also includes several functional viral proteins, including reverse transcriptase, integrase, and protease. The core is surrounded by capsid proteins; the nucleocapsid is surrounded by viral matrix protein. Complete virions are surrounded by an envelope derived from virus-modified host-cell cytoplasmic membranes; the envelope is studded by viral glycoproteins. The transmembrane protein extends from the matrix layer through the lipid bilayer to the external surface. The receptor-binding complex is anchored to the external portion of the transmembrane protein. Mature virions are spherical (~100 nm diameter).

The ssRNA genomes of retroviruses are similar to the host-cell mRNA. A repeat sequence is present at both ends of the ssRNA; the 5′ end is capped and the 3′ end polyadenylated. The order of sequences from the 5′ end to the 3′ end is Cap → Repeat sequence → unique sequence (U5) → the initiation site for initiation of minus-strand DNA synthesis → gag gene → pol gene → env gene → the initiation site for plus-strand DNA synthesis → a unique sequence (U3) → Repeat sequence → poly(A) sequence.

After entry into the cytoplasm of a susceptible cell, double-stranded DNA is synthesized by reverse transcription of both copies of the retroviral ssRNA. The viral-encoded DNA is transported into the nucleus, after which it is integrated into the host's genomic DNA. The process of forming new virions is initiated by transcription of the proviral DNA. The processed viral RNA is exported into the cytoplasm and genes for precursor viral proteins are translated. Virions are assembled at the cytoplasmic membrane and then released by budding; final virion maturation occurs by extracellular processing of viral proteins.

A characteristic of retroviruses is the high mutation rate and marked genomic heterogeneity of isolates. The major factors that contribute to this phenomenon include (1) error-prone reverse transcription, without proofreading correction, of the infecting virus genome; (2) recombination between the two genomic ssRNA strands during reverse transcription; and (3) the very high-level production of progeny viruses from infected cells.

Retroviruses are not assigned to a taxonomic order. The family *Retroviridae* has two subfamilies. The *Orthoretrovirinae* includes six genera, including *Deltaretrovirus* and *Lentivirus*. HTLV-1 is assigned the species name *Primate T-lymphotropic virus 1* in the *Deltaretrovirus* genus. Human immunodeficiency virus 1 and HIV-2 are named *Human immunodeficiency virus 1* (type species) and *Human immunodeficiency virus 2*, respectively, in the genus *Lentivirus* (Clavel et al. 1986b).

- *Human Immunodeficiency Viruses*: The human immunodeficiency viruses have a conical core surrounded by an envelope derived from viral-modified host-cell cytoplasmic membrane. Binding and entry of HIV into susceptible cells requires several specific receptors: CD4 (present on host helper T cells, CD4+ macrophages, and some dendritic cells) plus chemokine receptors, including CCR5 and CXCR4 (Klatzman et al. 1984; Simmons et al. 1998). The biological properties of HIV-1 isolates depend on the chemokine coreceptor(s) used by the virus (Berger et al. 1998). Isolates that exclusively use CXCR4 are T-cell tropic with rapid replication and syncytium formation. Isolates that use CCR5 exclusively are tropic to macrophages, replicate more slowly, and do not induce syncytium formation. Isolates that can use either CXCR4 or CCR5 have intermediate phenotypes.

  The *gag*, *pro*, *pol,* and *env* genes are translated from full-length mRNA transcripts of the proviral genome: *gag* and *env* in one reading frame and *pro* and *pol* from a second reading frame. In addition, several genes are transcribed from overlapping or unique reading frames, including several spliced gene products.

  Human immunodeficiency type 1 and 2 viruses evolved from simian viruses (Gao et al. 1999; Peeters et al. 1989; Daniel et al. 1985; Marx et al. 1991). These viruses may be distinguished by a number of characteristics, including clinical disease, specific antigens, and gene sequences (Clavel et al. 1986a). HIV-1 isolates may be further characterized into genetic groups and subtypes or clades (Wainberg 2004). Most HIV-1 isolates are in the M (main) group, which has a number of well-defined subgroups and recombinant forms with heterogeneous global distribution; clade B viruses are the predominant isolates in North America and Europe (Hemelaar et al. 2006; Osmanov et al. 2002). Group O (outlier) strains have mainly been isolated or acquired in Western Africa. Group N (non-M, non-O) and recombinant forms are also most commonly isolated from Western Africa.
- *Human T-Cell Leukemia Virus Type 1*: Mature HTLV virions have a spherical core, symmetrically placed within the envelope. The host-cell receptor is GLUT-1, a surface glucose transport molecule (Manel et al. 2003). The *gag*, *pro*, *pol,* and *env* genes are translated from full-length mRNA transcripts of the proviral genome: *gag* in one reading frame, *pro* and *env* from a second, and *pol* from a third reading frame. In addition, several spliced genes are transcribed from overlapping reading frames.

3.4 Summary

Recent and continuing progress to develop and use standardized and widely accepted methods for biological and taxonomic classification of viral pathogens has resulted in improvement in the medical response to viral illnesses. At a very basic level, these systems allow clinicians and scientists to communicate effectively and ensure the comparability of data generated by clinical or basic scientific studies. Further, accurate and standardized data is critical for understanding issues related to transmission, prevention, and treatment of viral illnesses. Establishing phylogenetic similarity to known viral pathogens may allow clinicians to anticipate the clinical behavior of new and emerging viral pathogens, as may be seen when virus mutation results in acquisition of new pathogenic mechanisms, like changes to antigens associated with evasion of the immune response of the host species or changes that allow a viral pathogen to jump from one species into new, susceptible species. As analytical tools improve, even more informative data relevant to clinical and pathologic characteristics of viral pathogens is anticipated.

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