Viral Gene Expression and Replication After uncoating and release of viral nucleoprotein into the cytoplasm, the viral genome is transported to sites of expression and replication. To produce infectious progeny, viruses must produce proteins necessary for replicating their nucleic acids as well as structural proteins necessary for coating their nucleic acids and for assembling nucleic acids and proteins into progeny virus. Different viruses use different strategies and gene repertoires to accomplish these goals. Most DNA viruses, except for poxviruses, replicate their nucleic acid and assemble into nucleocapsids in the cell nucleus. RNA viruses, except for influenza viruses, transcribe and replicate their RNA and assemble in the cytoplasm before envelopment at the cell plasma membrane. The replication strategies of DNA and RNA viruses and of positive- and negative-strand RNA viruses are presented and discussed separately below. Medically important viruses of each group are used for illustrative purposes.

POSITIVE-STRAND RNA VIRUSES RNA viruses of medical importance include positive-strand picornaviruses, flaviviruses, togaviruses, caliciviruses, and coronaviruses. Genome RNA from positive-strand RNA viruses is released into the cytoplasm without associated enzymes. Cell ribosomes recognize and associate with the viral genome's internal ribosome entry sequence and translate a virus-encoded polyprotein. Proteases within the polyprotein cleave out the viral RNA polymerase and other viral proteins necessary for replication. Antigenomic RNA is next transcribed from the genome RNA template. Positive-strand genomes and mRNAs are then transcribed from the antigenome RNA by the viral RNA polymerase and are translated into capsid proteins. Genomic RNA is encapsidated in the cytoplasm and released as the infected cell undergoes lysis.

NEGATIVE-STRAND RNA VIRUSES Medically important negative-strand RNA viruses include rhabdoviruses, filoviruses, paramyxoviruses, orthomyxoviruses, and bunyaviruses. The genomes of negative-strand viruses are frequently segmented. Negative-strand RNA viral genomes are released into the cytoplasm with an associated RNA polymerase and one or more polymerase accessory proteins. The viral RNA polymerase transcribes mRNAs as well as full-length antigenome RNA, which is the template for genome RNA replication. Viral mRNAs encode the viral RNA polymerase and accessory factors as well as viral structural proteins. Except for influenza virus, which transcribes its mRNAs and antigenome RNAs in the cell nucleus, negative-strand RNA viruses replicate entirely in the cytoplasm. All negative-strand RNA viruses, including influenza viruses, assemble in the cytoplasm.

DOUBLE-STRAND SEGMENTED RNA VIRUSES Double-strand RNA viruses are taxonomically grouped in the family Reoviridae. The medically important viruses in this group are rotaviruses and Colorado tick fever virus. Reovirus genomes have 10–12 RNA segments. Reovirus particles contain an RNA polymerase complex. These viruses replicate and assemble in the cell cytoplasm.

DNA VIRUSES Medically important DNA viruses include parvoviruses, which have small single-strand DNA genomes and cause transient arthritis, and polyomaviruses, including the smaller polyomaviruses such as JC virus, which causes progressive multifocal leukoencephalopathy in immunocompromised patients; BK virus; and Merkel cell polyomavirus. The larger HPVs cause warts as well as cervical, penile, and oral carcinomas. The next larger DNA viruses are adenoviruses, which mostly cause transient respiratory tract and ocular inflammatory disease. The herpesviruses include eight viruses that cause a wide range of inflammatory and malignant diseases in humans. EBV is an important cause of lymphomas and Hodgkin's disease in both immunocompromised and immunocompetent people and of nasopharyngeal carcinoma in southern Chinese and northern African populations. Cytomegalovirus (CMV) is an important cause of transplacental infections and neonatal neurologic impairment. Poxviruses, the largest DNA viruses and the largest viruses that infect humans (barely visible by light microscopy), cause smallpox, monkeypox, and molluscum contagiosum. Aside from those of poxviruses, other DNA virus genomes enter the cell nucleus and are transcribed by cellular RNA polymerase II.

After receptor binding and fusion with plasma membranes or endocytic vesicle membranes, herpesvirus nucleocapsids are released into the cytoplasm with tegument proteins and are transported along microtubules to a nuclear pore. Capsids then release DNA into the nucleus.

DNA virus transcription and mRNA processing depend on both viral and cellular proteins. For herpes simplex virus (HSV), a viral tegument protein enters the nucleus and activates immediate-early genes, the first genes expressed after infection. Transcription of immediate-early genes requires the viral tegument protein and cell transcription factors. HSV becomes nonreplicating, or latent, in neurons because essential cell transcription factors for expression of viral immediate-early genes are docked in the cytoplasm in neurons. Heat shock or other cell stresses can cause these cell factors to enter the nucleus, activate viral gene expression, and initiate replication. This information explains HSV-1 latency in neurons and activation of replicative infection.

For adenoviruses and herpesviruses, transcription of immediate-early genes results in expression of early proteins necessary for viral DNA replication. Viral DNA synthesis is required to turn on late-gene expression and production of viral structural components. The HPVs, polyomaviruses, and parvoviruses are not dependent on transactivators encoded from the viral genome for early-gene transcription. Instead, their early genes have upstream enhancing elements that bind cell transcription factors. The early genes encode proteins that are necessary for viral DNA synthesis and late-gene transcription. DNA virus late genes encode structural proteins necessary for viral assembly and for viral egress from the infected cell. Late-gene transcription is continuously dependent on DNA replication. Therefore, inhibitors of DNA replication also stop late-gene transcription.

Each DNA virus family uses unique mechanisms for replicating its DNA. Adenovirus and herpesvirus DNAs are linear in the virion. Adenovirus DNA remains linear in infected cells and replicates as a linear genome, using an initiator protein–DNA complex. In contrast, herpesvirus DNA circularizes in the infected cell, and genomes replicate into linear concatemers through a "rolling-circle" mechanism. Full-length DNA genomes are cleaved and packaged into virus. Herpesviruses encode a DNA polymerase and at least six other viral proteins necessary for viral DNA replication. Acyclovir and ganciclovir prevent viral DNA synthesis when they are phosphorylated and incorporated into DNA by the viral polymerase. Herpesviruses also encode enzymes that increase the deoxynucleotide triphosphate pools. HPV and polyomavirus DNAs are circular both within the virus and in infected cells. These genomes are reproduced by cellular DNA replication enzymes and remain circular through replication and packaging. HPV and polyomavirus early proteins are necessary for DNA replication in both latent and viral replicative phases. Early viral proteins stimulate cells to remain in cycle, facilitating viral DNA replication.

Parvoviruses have negative single-strand DNA genomes and are the smallest DNA viruses. Their genomes are half the size of HPV genomes and include only two genes. The replication of autonomous parvoviruses, such as B19, depends on cellular DNA replication and requires the virus-encoded Rep protein. Other parvoviruses, such as adeno-associated virus (AAV), are not autonomous and require helper viruses of the adenovirus or herpesvirus family for their replication. AAV is being used as a potentially safe human gene therapy vector because its replication protein causes integration at a single chromosome site. The small genome size limits the range of proteins that can be expressed from AAV vectors.

As stated above, poxviruses are the largest DNA viruses. They are unique among DNA viruses in replicating and assembling in the cytoplasm. To accomplish cytoplasmic replication, poxviruses encode transcription factors, an RNA polymerase II orthologue, enzymes for RNA capping, enzymes for RNA polyadenylation, and enzymes for viral DNA synthesis. Poxvirus DNA also has a unique structure. The double-strand linear DNA is covalently linked at the ends, making a covalently closed double-strand circular genome. Replication of the circular genomes is initiated by nicking in inverted repeats at the ends of the linear DNA. During DNA replication, the genome is cleaved