RESISTANCE TO VIRAL INFECTIONS

Resistance to viral infections is initially provided by factors that are not virus-specific. Physical protection is afforded by the cornified layers of the skin and by mucous secretions that continuously sweep over mucosal surfaces. Once the first cell is infected, IFNs are induced and confer resistance to RNA virus replication. Viral infection may also trigger the release of other cytokines from infected cells. These cytokines may be chemotactic to inflammatory and immune cells. Viral protein epitopes expressed on the cell surface in the context of MHC class I and II proteins can stimulate the expansion of T cell populations with receptors that can recognize virus-encoded peptides presented on the cell surface by MHC class I proteins. Cytokines and antigens released by virus-induced cell death further attract inflammatory cells, dendritic cells, granulocytes, natural killer (NK) cells, and B lymphocytes to sites of infection and to draining lymph nodes. IFNs and NK cells are particularly important in containing viral infection for the first several days. Granulocytes and macrophages are also important in the phagocytosis and degradation of viruses, especially after an initial antibody response.

By 7–10 days after infection, virus-specific antibody responses, virus-specific human leukocyte antigen (HLA) class II–restricted CD4+ helper T lymphocyte responses, and virus-specific HLA class I–restricted CD8+ cytotoxic T lymphocyte responses develop. These responses, whose magnitude typically increases over the second and third weeks of infection, are important for rapid recovery. Also between the second and third weeks, the antibody type usually changes from IgM to IgG; IgG or IgA antibody can then be detected at infected mucosal surfaces. Antibody may directly neutralize virus by binding to its surface and preventing cell attachment or penetration. Complement can significantly enhance antibody-mediated virus neutralization. Antibody and complement can also lyse virus-infected cells that express viral membrane proteins on the cell surface. Cells infected with a replicating enveloped virus usually express the virus-envelope glycoproteins on the cell plasma membrane. Specific antibodies can bind to the glycoproteins, fix complement, and lyse the infected cell.

Antibody and CD4+/CD8+ T lymphocyte responses to viral infection can remain at high levels for several months after primary infection but usually wane over time. Low-level persistence of antibody-producing B lymphocytes and CD4+ or CD8+ T lymphocyte responses as memory cells can provide a rapid response to a second infection or an early barrier to reinfection with the same virus. Redevelopment of T cell immunity may take longer than secondary antibody responses, particularly when many years have elapsed between primary infection and reexposure. However, persistent infections or frequent reactivations from latency can result in sustained high-level T cell responses. EBV and CMV typically induce high-level CD4+ and CD8+ T cell responses that are maintained for decades after primary infection.

Some viruses have genes that alter innate and acquired host defenses. Adenoviruses encode small RNAs that inhibit IFN-induced, protein kinase R (PKR)–mediated shutoff of infected-cell protein synthesis. Adenovirus E1A can also directly inhibit IFN-mediated changes in cell gene transcription. Moreover, adenovirus E3 proteins prevent tumor necrosis factor (TNF)–induced cytolysis and block HLA class I synthesis by the infected cell. HSV ICP47 and CMV US11 also block class I antigen presentation. EBV encodes an interleukin (IL) 10 homologue that inhibits NK and T cell responses. Vaccinia virus encodes a soluble receptor for IFN-α and binding proteins for IFN-γ, IL-1, IL-18, and TNF, which inhibit host innate and adaptive immune responses. Vaccinia virus also encodes a caspase inhibitor that inhibits the ability of CD8+ cytotoxic T cells to kill virus-infected cells. Some poxviruses and herpesviruses encode chemokine-binding proteins that inhibit cell inflammatory responses. The adoption of these strategies by viruses highlights the importance of the corresponding host resistance factors in containing viral infection and the importance of redundancy in host resistance.

The host inflammatory and immune responses to viral infection do not come without a price. These responses contribute to the symptoms, signs, and other pathophysiologic manifestations of viral infection. Inflammation at sites of viral infection can subvert an effective immune response and induce tissue death and dysfunction. Moreover, immune responses to viral infection could, in principle, result in immune attack upon cross-reactive epitopes on normal cells, with consequent autoimmunity.

INTERFERONS

All human cells can synthesize IFN-α or IFN-β in response to viral infection. These IFN responses are usually induced by the presence of double-strand viral RNA, which can be made by both RNA and DNA viruses and sensed by double-strand RNA binding proteins (e.g., PKR and RIG-I) in the cell cytoplasm. IFN-γ is not closely related to IFN-α or IFN-β and is produced mainly by NK cells and by immune T lymphocytes responding to IL-12. IFN-α and -β bind to the IFN-α receptor, whereas IFN-γ binds to a different but related receptor. Both receptors signal through receptor-associated JAK kinases and other cytoplasmic proteins, including "STAT" proteins, which are tyrosine-phosphorylated by JAK kinases, translocate to the nucleus, and activate promoters for specific cell genes. Three types of antiviral effects are induced by IFN at the transcriptional level. The first effect is attributable to the induction of 2′-5′ oligo(A) synthetases, which require double-strand RNA for their activation. Activated synthetase polymerizes oligo(A) and thereby activates RNAse L, which in turn degrades single-strand RNA. A second effect results from the induction of PKR, a serine and threonine kinase that is also activated by double-strand RNA. PKR phosphorylates and negatively regulates the translational initiation factor eIF2α, shutting down protein synthesis in the infected cell. A third effect is initiated through the induction of Mx proteins, a family of GTPases that is particularly important in inhibiting the replication of influenza virus and vesicular stomatitis virus. These IFN effects are mostly directed against the infected cell, causing virus and cell dysfunction and thereby limiting viral replication.

DIAGNOSTIC VIROLOGY

A wide variety of methods are used to diagnose viral infection. Serology and virus isolation in tissue culture remain important standards. Acute- and convalescent-phase sera with rising titers of antibody to virus-specific antigens and a shift from IgM to IgG antibodies are generally accepted as diagnostic of acute viral infection. Serologic diagnosis is based on a more than fourfold rise in IgG antibody concentration when acute- and convalescent-phase serum samples are analyzed at the same time.

Immunofluorescence, hemadsorption, and hemagglutination assays for antiviral antibodies are labor-intensive and have been replaced by enzyme-linked immunosorbent assays (ELISAs), which generally use the specific viral proteins most frequently targeted by the antibody response. The proteins are purified from virus-infected cells or produced by recombinant DNA technology and are attached to a solid phase, where they can be incubated with serum, washed to eliminate nonspecific antibodies, and allowed to react with an enzyme-linked reagent to detect human IgG or IgM antibody specifically adhering to the viral antigen. The amount of antibody can then be quantitated by the intensity of a color reaction mediated by the linked enzyme. ELISAs can be sensitive and automated. Western blots can simultaneously confirm the presence of antibody to multiple specific viral proteins. The proteins are separated by size and transferred to an inert membrane, where they are incubated with serum antibodies. Western blots have an internal specificity control because the level of reactivity for viral proteins can be compared with that for cellular proteins in the same sample. Western blots require individual evaluation and are inherently difficult to quantitate or automate.

Isolation of virus in tissue culture depends on infection and replication in susceptible cells. Growth of virus in cell cultures can frequently be identified by effects on cell morphology under light microscopy. For example, HSV produces a typical cytopathic effect in rabbit kidney cells within 3 days. Other viral cytopathic effects may not be as diagnostically distinctive. Identification usually requires confirmation by staining with virus-specific monoclonal antibodies. The efficiency and speed of virus identification can be enhanced by combining short-term culture with immune detection. In assays with "shell vials"