Viral Pathogenesis

- **Pathogenesis:** the process by which one organism causes disease in another

- Two components of viral disease:
  - Effects of virus replication on the host
  - Effects of host response on virus and the host

- The goal of studies on pathogenesis is to identify the viral and host genes that influence the production of
Respiratory tract

- Most common route of viral entry
- Absorptive area of lung: 140 m²; ventilation rate 6 L/min
- Barriers to infection: swallowing; ciliary action from lower tract; macrophages in alveoli (no cilia or mucus); IgA
- Viruses enter by aerosolized droplets from cough or sneeze, or contact with saliva
- Large droplets lodge in nose; smaller in airways or alveoli
Alimentary tract

• Eating, drinking, social activities introduce viruses into the alimentary tract
• Designed to mix, digest, absorb food, so contents are always in motion; good opportunities for virus-cell interactions
• Extremely hostile environment: stomach is acidic, intestine is alkaline; presence of digestive enzymes, bile detergents, mucus, antibodies, phagocytic cells
• Viruses have evolved to infect are resistant: enteroviruses; reovirus (require proteases); enteric coronavirus (enveloped!)
Urogenital tract

- Protected by mucus, low pH
- Minute abrasions from sexual activity may allow viruses to enter
- Some viruses produce local lesions (HPV)
- Some viruses spread from urogenital tract (HIV, HSV)

Eye

- Sclera and conjunctiva are route of entry
- Every few seconds eyelid passes over sclera, washing away foreign particles; little opportunity for infection
- Infection usually occurs after injury: grit, ophthalmologic procedures, improperly sanitized swimming pools
- Localized infection: conjunctivitis
- Disseminated infection: EV70 spread to CNS
- HSV-1 can infect cornea, blindness may result, virus spread to sensory ganglia
Skin

- Outer layer of dead, keratinized cells cannot support viral infection; entry usually occurs by breaks or punctures
- Skin abrasions; insect or animal bites; needle punctures
- Epidermis is devoid of blood or lymphatics; local replication
- Dermis and sub-dermal tissues are highly vascularized; infection may spread

Viral Spread

- After replication at the site of entry, viruses may remain localized: virus spreads within the epithelium and is contained by tissue structure and immune system
- Some viruses spread beyond the primary site: disseminated; if many organs are infected, systemic
- Physical and immune barriers must be breached
Viral Spread

- Below the epithelium is the basement membrane; integrity can be compromised by epithelial inflammation and destruction.
- Below basement membrane are subepithelial tissues, where virus encounters tissue fluids, lymphatic system, and phagocytes; all play roles in clearing and spreading infection.
- Role of directional release of virus from the basal epithelium:
  - Apical release facilitates virus dispersal; virus usually does not invade underlying tissues.
  - Basolateral release provides access to underlying tissues and may facilitate systemic spread.
  - Sendai virus: apical release from respiratory tract, local infection; mutant that is released from both apical and basal surfaces causes disseminated infection.
Hematogenous Spread

- Viruses that produce disseminated infection often do so by entering the blood
- Viruses may enter blood directly through capillaries, by replicating in endothelial cells, or through vector bite
- Virus in the extracellular fluids is taken up by lymphatic capillaries, which are more permeable than circulatory capillaries, then spread to blood
- Once in blood, virus has access to almost every tissue
- In lymph nodes, viruses encounter lymphocytes and other immune cells, and may replicate in them; may also spread infection to distant tissues
- Other viruses spread freely in the blood

Viremia

- Presence of infectious virus in the blood
- Active viremia: results from virus replication
- Passive viremia: results from virus introduced into the blood without replication
- Diagnostic value
- Practical problems (blood supply)
Pathogenesis of mousepox

- Frank Fenner

- First to demonstrate how disseminated viral infections develop from local multiplication to primary and secondary viremia to target organs

Neural spread

- Many viruses spread from primary site of infection by entering local nerve endings
- For some viruses (rabies, alpha herpesviruses) neural spread is definitive characteristic of pathogenesis
- For other viruses (poliovirus, reovirus) invasion of the CNS is an infrequent diversion from normal replication and hematogenous spread
Viral spread to the central nervous system

<table>
<thead>
<tr>
<th>Pathway</th>
<th>Viruses</th>
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<tbody>
<tr>
<td>Neural</td>
<td>Poliovirus, yellow fever virus, mouse hepatitis virus, Venezuelan encephalitis virus, rabies virus, reovirus (type 3 only: type 1 spread by viremia), herpes simplex virus types 1 and 2, pseudorabies virus</td>
</tr>
<tr>
<td>Olfactory</td>
<td>Poliovirus (experimental), herpes simplex virus, coronavirus</td>
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<tr>
<td>Hematogenous</td>
<td>Poliovirus, coxsackievirus, arenavirus, mumps virus, measles virus, herpes simplex virus, cytomegalovirus</td>
</tr>
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</table>

Infections of the CNS

- A **neurotropic** virus can infect neural cells; infection may occur by neural or hematogenous spread from a peripheral site
- A **neuroinvasive** virus can enter the CNS after infection of a peripheral site
- A **neurovirulent** virus can cause disease of nervous tissue
  - HSV: low neuroinvasiveness, high neurovirulence
  - Mumps: high neuroinvasiveness, low neurovirulence
  - Rabies: high neuroinvasiveness, high neurovirulence
Tissue invasion

Liver, spleen, bone marrow, adrenal glands
Renal glomerulus, pancreas, ileum, colon
CNS, connective tissue, skeletal & cardiac muscle

Tissue invasion: Liver

Hepatitis B virus
Kupffer cells
Growth in Kupffer cell
Inactivation in Kupffer cell
Passage through Kupffer cell
± hepatic cell growth
± bile duct excretion
No uptake
Tissue invasion: blood-brain junction

- Replication in endothelial cells
- Transcytosis
- Trafficking lymphocyte or monocyte
- Basement membrane
- Endothelial cell
- Capillary lumen

Tissue invasion: CNS

- Blood vessel in choroid plexus
- Ventricles
- Meninges
- Plexus
- Brain parenchyma
- CSF
- Nerve
- From peripheral nerve ending or nasal mucosa
- Cerebral blood vessels
- Meningeal blood vessels
Tissue Tropism

• The spectrum of tissues infected by a virus
  — e.g. an enteric virus replicates in the gut and not in the lung; a neurotropic virus replicates in cells of the nervous system and not in hematopoietic cells

• The tropism of some viruses is limited; other viruses are pantropic, e.g. can replicate in many organs

• What are the determinants of viral tropism?

Determinants of Tissue Tropism

• Cell receptors for viruses
  — e.g. HIV-1 & CD4; EBV & CR2 but not poliovirus or influenza virus

• Cellular proteins that regulate viral transcription
  — e.g. JC papovavirus replicates in oligodendrocytes because the viral enhancer is active only in this cell type

• Cell proteases
  — e.g. cleavage of influenza virus HA by serine proteases
Viral virulence

- The capacity of a virus to cause disease in an infected host
- A virulent virus causes significant disease, while an avirulent or attenuated virus causes reduced or no disease
- Virulence can be quantitated:
  - LD$_{50}$ (Lethal Dose 50%; amount of virus needed to kill 50% of infected animals)
  - The mean time to death
  - The mean time to appearance of symptoms
  - Measurement of fever, or weight loss
  - Measurement of pathological lesions (poliovirus); reduction in blood CD4+ lymphocytes (HIV-1)
What makes viruses virulent?

- A major goal of virology is to identify viral and host genes that determine virulence.
- Virulence genes are usually identified by mutation: deletion or disruption of one of these genes results in a virus that causes reduced or no disease in a specified system.
- Viral genes affecting virulence fall into four classes:
  - Those that affect the ability of the virus to replicate
  - Those that modify the host’s defense mechanisms
  - Those that enable the virus to spread in the host
  - Those which have intrinsic cell killing effects

![Diagram of virus phenotypes](image)
Genes that modify the host’s defense mechanisms

- Virokines (secreted proteins that mimic cytokines, growth factors, or similar extracellular immune regulators) and viroceptors (homologs of host receptors or cell surface immune molecules)
- Mimic normal cellular molecules critical to host defense
  - sabotage the body’s innate and adaptive defenses
  - Not required for growth in cell culture
  - Most have been found in large DNA viruses (pox, herpes, adenovirus)
- Examples:
  - Soluble cytokine receptor - bind cytokines, block action
  - Proteins that bind key proteins in complement cascade
  - Proteins that affect MHC-1 antigen presentation

A herpesvirus gene that binds cytokines: effect on survival in mice

![Graph showing survival rates of different models of virus infection]
**Toxic viral proteins**

- NSP4 nonstructural glycoprotein of rotaviruses: a viral enterotoxin
- When expressed in cells, causes increase in intracellular calcium.
- When fed to young mice, causes diarrhea by potentiating chloride secretion. Thus, NSP4 triggers a signal transduction pathway in intestinal mucosa

**How do viruses injure cells?**

- Infection of cultured cells by cytolytic viruses: cytopathic effects
- Many viruses cause inhibition of host protein and RNA synthesis, which leads to loss of membrane integrity, leakage of enzymes from lysosomes, cytoplasmic degradation
- Syncytium formation by enveloped viruses (parainfluenza, HIV)
- Virus infection can induce *apoptosis* (programmed cell death)
Mechanisms of cell injury by viruses

Non-cytolytic viruses: disease usually a consequence of the immune response: immunopathology

<table>
<thead>
<tr>
<th>Proposed mechanism</th>
<th>Virus</th>
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<tr>
<td>CD8⁺ T cell mediated</td>
<td>- Coxackievirus B</td>
</tr>
<tr>
<td></td>
<td>- Lymphocytic choriomeningitis virus</td>
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<td>- Sth Nombre virus</td>
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<td></td>
<td>- Human immunodeficiency virus type 1</td>
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<td></td>
<td>- Hepatitis B virus</td>
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<tr>
<td>CD4⁺ T cell mediated</td>
<td>- Theiler's virus</td>
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<tr>
<td>Th1</td>
<td>- Mouse coronavirus</td>
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<td>- Semliki Forest virus</td>
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<td>- Measles virus</td>
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<td>- Virea virus</td>
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<td>- Herpes simplex virus</td>
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<td>Th2</td>
<td>- Respiratory syncytial virus</td>
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<tr>
<td>Antibody mediated</td>
<td>- Dengue virus</td>
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<td></td>
<td>- Feline infectious peritonitis virus</td>
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- Lesions associated with CD8⁺ T cells: myocarditis caused by coxsackievirus B
- Hypothesis: tissue damage due to cytotoxicity of CD8⁺ T cells; perforin knockout mice develop less severe disease
- CD8⁺ T cells may also release proteins that recruit inflammatory cells which elaborate proinflammatory cytokines

A

B
Lesions associated with B cells: Dengue

- Caused by Dengue virus, transmitted mainly by bites of *Aedes aegypti* mosquitoes
- Endemic in the Caribbean, Central and South America, Africa and Southeast Asia
- 50 million infections/year
- Primary infection is usually asymptomatic, but may result in standard symptoms of virus infection: *acute febrile illness with severe headache, back and limb pain and rash*. Severe aches and pains in the bones.
  - Normally self-limiting, patients recover in 7-10 days

Dengue Fever

- In 1/14,000 *primary* infections, people get *Dengue Hemorrhagic Fever (DHF)*, a life threatening disease.
- Patients produce antibodies to virus, but there are four serotypes, and no cross-protection
- Non-protective antibodies can *enhance* the infection of peripheral blood monocytes by *Fc-receptor mediated uptake* of antibody coated virus particles. Infected macrophages release cytokines, causing severe symptoms
- After *secondary* dengue infections, (i.e. infections of people with antibody to Dengue virus), the incidence of DHF 1/90.
Cell injury associated with free radicals

- Superoxide (O$_2^-$) and nitric oxide (NO) are produced during the inflammatory response.

- NO is made by nitric oxide synthase, an interferon-inducible enzyme.

- Low concentrations of NO have a protective effect, high concentrations cause tissue damage by reacting with O$_2^-$ to form peroxynitrite, which is much more reactive than either radical.