Viral Pathogenesis → Rapid replication, frequent mutation, ability to encode virulence genes
- Viruses are **obligate intracellular parasites** that require a host for replication

4 Patterns of Viral Disease:
1. **Acute** Infection → Transient replication of virus in target cells, with brief plasma viremia (e.g. *rhinovirus*, *rotovirus*, *influenza virus*)
2. **Persistent** Infection → Replication in host cells continues over time after initial infection
   a. Host is unable to develop effective control measures
   b. Eventual overt plasma viremia leads to host death (**lymphocytic choriomeningitis**)
3. **Latent Reactivating** Infection → Replication of virus in cells is sporadic and intermittent
   a. Infection isn’t cleared between bursts; virus remains dormant-latent in host (**herpes**)
4. **Slow-virus** Infection → An initial acute/primary phase infection is followed by semi-effective host response which prohibits virus replication until a later time (**HIV/AIDS**)

**Virulence** is under polygenic control
1. Ability of virus to replicate
2. Defeat the host’s defense mechanisms
3. Promote virus spread within and among hosts → **virokines** (mimic host cytokines) and **viroreceptors** (mimic host cell receptors to sequester cytokines)
4. Gene products that are directly toxic to the host (**virotoxins**)

**Tropism** = the particular cell type/organ infected by a virus
- **Neurotropic** = CNS; **pneumotrophic** = respiratory tract; **enterotropic** = GI tract; **pantropic** = multiple body systems
- Viral receptors are required for viral entry, determine host tropism
  o Some viruses need multiple receptors or a co-receptor
  o Examples = integrins, Ig-like molecules, glycosaminoglycans, CHO

Viral Spread → systemic spread must cross barriers like the basement membrane
- **Directional release** is a major determinant of infection pattern
- Blood flow allows spread throughout body, either freely in plasma or within blood cells

Innate host defenses are aided by physical barriers to viral infection:
- Respiratory tract → mucociliary apparatus, alveolar macrophages, adaptive immune response
- GI Tract → Low stomach pH, digestive enzymes, flow, adaptive immune response
- Skin → Thickness of epidermis, skin oils; little circulation prevents systemic infections

Host susceptibility to viral disease depends on genetics (MHC class I gene repertoire) as well as non-genetic determinants (elderly, male, pregnancy, IC pts, malnutrition, stress, steroid use, smoking)

Types of Viral Damage to Tissues
1. **Cell Damage** → Swelling, necrosis, and apoptosis
   a. Rotaviruses ↑ Cl secretion → osmotic diarrhea and enterocyte swelling
   b. **Necrosis** histology = ballooning degeneration, clumping of chromatin
   c. Apoptosis may aid viral spread in some viruses, but be inhibited by others to establish latency/long-term viral replication
2. **Inclusion Bodies** → classic morphology/histology hallmark of viral infection
   a. Vary widely in appearance → intranuclear and/or intracytoplasmic; stain different colors
3. **Syncytia**/multi-nucleated cells → caused by fusion of epithelial cells or macrophages
   - Some viruses induce fusion of cell membranes, possibly to help cell to cell spread
4. **Cellular hyperplasia/proliferation** → generally self-limiting and transient
   - May also be pre-neoplastic, and neoplasia may develop (EBV and nasopharyngeal CA; HPV and cervical CA)

The most common/classic host inflammatory responses to a viral infection = **mononuclear infiltrates** made up of lymphocytes, plasma cells, and macrophages

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**Introduction to Virology**

**Virus Characteristics**
- Smallest infectious agent (20-300nm), containing either DNA or RNA
- Surrounded by a **capsid** (made up of capsomeres)
- Some also surrounded by **lipid envelope** (makes more susceptible to heat and chemicals, but allows host to recognize virus as self); naked virus = no envelope
- Origin Hypotheses
  - Derived from RNA or DNA components of host cells
  - Degenerate forms of intracellular parasites
- **Virion** = minimum amount of virus that makes it infectious (RNA only, RNA + capsid, etc.)

Viruses are classified by: 1.) Nucleic acid in the virion, 2.) Symmetry in the capsid, 3.) Presence/absence of envelope, and 4.) Dimensions of virion and capsid (**icosahedral or helical**)

**Viral Genome Structure**
- **RNA** → Stay in cytoplasm, bring enzymes for replication/transcription (potential target for tx)
  - Single-stranded
    - **Plus strand** RNA = mRNA (uses host ribosomes in cytoplasm)
    - **Minus strand** RNA (segmented and non-segmented)
  - Double-stranded (segmented)
- **DNA** → In nucleus, use eukaryotic cell replication machinery (DNA pol) → no good target for tx
  - Only exception = herpes virus (future lectures)

**Viral replication cycle (LYTIC example):**
Attachment → penetration → uncoating → transcription of early mRNA → translation of early proteins → replication of viral DNA → transcription of late mRNA → translation of late proteins → virion assembly → release (LYTIC)

**Nonstructural** proteins = early proteins → encoded by viral genome, not packaged into virion, usually enzymes or transcription factors necessary for viral replication within host cell
**Structural proteins** = late proteins → encoded by viral genome, packaged into virion

Different viruses use different cell surface molecules for attachment: (only memorize for HIV)
- **Carbohydrates** (linked to proteins/lipids → sialic acid, glycosaminoglycans)
- **Lipids** (glycolipids, proteolipids)
- **Proteins** (Ig superfamily, complement regulatory proteins, integrins, TNF receptor superfamily)
Viruses can evolve by:

1. **Mutation** → RNA Viruses evolve rapidly because they produce a large # of progeny and RNA polymerases lack proofreading capability → creation of “quasispecies”
2. **Recombination** → 2 similar viruses infect same cell, and genomes combine (“antigenic shift” in Influenza causes epidemics/pandemics and fatal flu strains)
3. **Reassortment** → Viruses with segmented genomes create small changes (“drifts” in Influenza create the variations each flu season)

Viruses overcome limitations of a small genome by pre- and post-translational modifications

Types of viral infections = acute, **persistent** (e.g. HIV, HCV → continuous replication), and **latent** infections (e.g. herpes → periods of dormancy)

- Persistence requires not killing the host or host cells, not being eliminated by host response
- Incubation periods important to determine cause (days v weeks v years)

In addition to innate and adaptive immunity, there are host-specific/genetic defenses against viruses:

- **CCR5-Δ32** chemokine in 10% of Europeans = protective against HIV
- **Fut-2** enzyme mutation in epithelia cells = protective against diarrhea w/ norovirus infection
- **Interferons** are part of the innate response → induced by DS RNA; nonspecific ↓ in protein synthesis in nearby non-infected cells to inhibit translation of viral mRNA
  - Used to treat viral infections HBV/HCV, but has lots of side effects (malaise, fatigue)

An effective response eliminates infectious virus from blood/fluids (prevent spread), from tissue (“cure”) and provides immunity against re-infection

- Cell-mediated contribution = clear infected cells, activate macrophages, B cell help to make Ig

**Serological tests** diagnose viral infections (enzyme immunoassay, radioimmunoassay, western blot)

**Biological activity** gives information on function of the antibody (neutralization, complement fixation, hemagglutination inhibition)

- Neutralizing Ab’s work outside and inside the cell (block attachment, penetration, uncoating)

**Public Health Impact** → seasonal influenza, HIV is #1 killer of young women, HBV/HCV is #1 cause of hepatocellular cancer and liver transplants, 165,000 measles deaths and ½ million rotovirus deaths annually

What to know for virology:

* Don’t memorize classifications of viruses we learn; only know steps in viral life cycle that are important in chemotherapy treatments → **know** retroviral life cycle (HIV)
* Most cells have **lytic** life cycles (release virion) → viruses with lysogenic life cycles integrate into host genome (most chronic viruses → herpes, EBV, HIV, hepatitis)
* Memorize the transmission methods for every virus
Parvovirus Infections
- Bocavirus → associated with respiratory disease in infants (don’t need to know, no tx)
- Erythrovirus (Parvovirus B19) → Erythema infectiosum (fifth disease), arthritis, aplastic anemia, chronic anemia

Replication → Very small genome, insufficient coding capacity for replication; must replicate in a host cell in S phase to use host DNA replication enzymes

Epidemiology → Human virus (no other reservoir), respiratory transmission (or by transfusion)
- 70% adults infected by age 50 → only infected once, neutralizing Ig prevents re-infection

Pathogenesis: Entrance via respiratory epithelium → viremia → skin, bone marrow symptoms
- Binds to P group antigen on erythroid cell surfaces (RBC precursors), replicates, and lyses
- In adults, manifests in bone marrow; in fetus, erythroid cells found in liver, marrow, other organs
  - Can be transmitted vertically to fetus if mother is infected

Clinical presentation of parvovirus B19 depends on patient:
- Healthy adults (RBCs live ~60 days) → no big change in RBCs before virus is cleared by Ig
  - Antibodies binding virus deposit in joints and skin, causing arthritis and rash (“slap cheek”)
- Patients with disorders of RBCs (shorter life in circulation), will see transient anemia
- Immunodeficient patients (no Ig) → pure red cell aplasia/anemia (NO RBC’s left)
  - Tx: Must treat with transfusions and IV Ig
- In fetus → severe anemia leading to hydrops fetalis (heart hypertrophy & failure, edema, death)
  - Particularly occurs <20 weeks of gestation
  - Tx: Can transfuse fetus before birth, but survival leads to immune tolerance and persistence of virus (congenital RBC aplasia)

Summary of Parvovirus B19-Induced diseases *know this chart

<table>
<thead>
<tr>
<th>Syndrome</th>
<th>Clinical Features</th>
<th>Host Characteristic</th>
</tr>
</thead>
<tbody>
<tr>
<td>Fifth Disease</td>
<td>Rash, arthralgia/arthritis</td>
<td>Children and adults</td>
</tr>
<tr>
<td>Transient aplastic crisis</td>
<td>Severe acute anemia</td>
<td>Hemolysis (e.g. Sickle Cell)</td>
</tr>
<tr>
<td>Pure red cell aplasia</td>
<td>Chronic anemia</td>
<td>Immunodeficiency</td>
</tr>
<tr>
<td>Hydrops fetalis</td>
<td>Fatal anemia, heart failure</td>
<td>Mid-trimester fetus</td>
</tr>
<tr>
<td>Congenital red cell aplasia</td>
<td>Non-regenerative, chronic anemia</td>
<td>Treated hydrops fetalis</td>
</tr>
</tbody>
</table>

Papillomaviruses → many types (some hands/feet), divided into 2 depending on risk of causing cancer:
1. High Risk (e.g. 16 and 18; causes warts → dysplasia → GU/anal/pharyngeal CA)
2. Low Risk (e.g. 6 and 11; causes 90% genital warts → low-grade dysplasia/NO CA)

All cervical carcinomas are caused by HPV → HPV-16 and -18 account for 70% invasive cervical CA
- Initial infection is common, asymptomatic and cleared within a few months
- Single biggest risk factor for progression of high risk strains to carcinoma is HPV persistence
- Other risk factors → age, infection with multiple HPV types, smoking, immunosuppression
  - HIV+ patients at higher risk → cervical CA is an AIDS-defining diagnosis

Transmission → sexual contact, virus found on GU tract, anal area, hands (men)
- Viral entry into body via micro-trauma abrasions → enters the basal layer of the epidermis
- Basal layer of epidermis has high rates of replication, DNA virus takes advantage
- Peak incidence in 19-24 year olds; 40% females infected with only 1 partner
Progressive development of cervical carcinoma (development of persistent/latent infection)

- Initial infection usually cleared in 6-9 months by immune response
  - Virus released from cell 3 weeks after initial infection
- Low-grade dysplastic (pre-cancerous) changes can occur if infection is not cleared within this time
  - Immune system can still clear low-grade dysplastic changes
- If not removed, progresses to higher grade dysplasia → carcinoma in situ → invasive cervical cancer (1-20 years)

Important Early Proteins to Know in HPV Infection:

- **E2** → controls replication, keeps replication at a low level
- **E6 and E7** → suppress tumor suppressor genes; allowing immortalization of cells/CA formation
  - DNA integrates into host cell near E2 gene, disrupting E2 function
  - No slowing of E6/7 → unchecked growth → warts → gradual CA development
    - Need BOTH E6 and E7 for host cell immortalization
Cervical CA = #1 cause of female CA deaths in developing countries (caught in US by routine pap smears)

Low Risk HPV Types 6 and 11 mainly cause warts and don’t lead to cancer

**Exception** = infants born to moms with low-risk infection can develop warts in respiratory tract, which must be surgically removed; more likely to get respiratory tract cancers 40-50 years later

HPV Vaccine → Includes capsid (main structural/late protein L1) without DNA

- Bivalent vaccine → Only protective against types 16 and 18 (70% cervical CA, several others)
- Tetravalent vaccine → 4 types (16, 18, 6, 11) → includes protection against low risk warts in order to market to men (simultaneously protect women from transmission from vaccinated men)
  - Won’t prevent CA if already infected, but will prevent initial infection

**SUMMARY**

<table>
<thead>
<tr>
<th>Parvoviruses/B19</th>
<th>Papillomaviruses/HPV</th>
</tr>
</thead>
<tbody>
<tr>
<td>- Small, single-stranded DNA, no envelope</td>
<td>- Double-stranded DNA, circular genome, no envelope</td>
</tr>
<tr>
<td>- Dependent on helper virus or enzymes of dividing cell for DNA replication (induce S phase)</td>
<td>- Causes warts</td>
</tr>
<tr>
<td>- B19 causes rash, anemia, and arthritis</td>
<td>- High risk types transform cells, predispose to CA</td>
</tr>
<tr>
<td></td>
<td>- E6 &amp; E7 disrupt cell cycle regulation (E2)</td>
</tr>
<tr>
<td></td>
<td>- 2 vaccines have been developed</td>
</tr>
</tbody>
</table>

Other Medically Important Viruses:

Seasonal variation of respiratory infections gives clue to causative agent **will be a question on exam**

- Adenovirus → Winter
- Enterovirus (e.g. polio) → summer/fall
- Rhinovirus → any time of year
- Para-Influenza Viruses (PIV) → Summer, fall, winter
- Influenza → Winter
- Respiratory Syncytial Virus (e.g. RSV, influenza) → winter

**Single Stranded DNA Viruses:** Parvovirus

**Double Stranded DNA Viruses:** Adenoviruses, Papillomaviruses, Polyomaviruses, Herpesviruses, Hep B
**Adenovirus** \(\rightarrow\) ds DNA, non-enveloped; 49 types cause human infection
- All types are transmitted by direct contact, small droplet, fecal-oral transmission, occasionally waterborne transmission
  - Some types capable to establishing persistent asymptomatic infections
- Clinical manifestations = respiratory infections (cold, bronchitis, pneumonia, ARDS), gastroenteritis, conjunctivitis, cystitis (often hemorrhagic)
- Host risk factors = SMI defects at ↑‘d risk of severe infections
- Treatment = supportive (can’t target much on DNA viruses)

**Single Stranded (Plus strand) RNA Viruses**

**Picornavirus** \(\rightarrow\) non-enveloped, + strand, small (“pico”) RNA viruses; genomic RNA is infectious
- Includes rhinoviruses, enteroviruses, and hepatoviruses (Hep A)

**Rhinovirus** \(\rightarrow\) Year-round infections, but peaks in early fall and late spring
- Common cause of upper airway infections, “common cold”, >100 serotypes
- Adapted to grow in the nasal passages (cooler temps)
- Transmission via respiratory route; treatment is supportive

**Enterovirus** \(\rightarrow\) Replicate in enteric tract but do not necessarily cause GI symptoms (coxsackie viruses, numbered enteroviruses and polioviruses)
- Infections in summer and early fall
- Replication in intestine → local lymph nodes → viremia → CNS, muscle, and skin
  - Skin (coxsackie) = hand-foot-mouth disease, mostly in kids
  - Muscle/heart (coxsackie A/B, echovirus) = pericarditis, myocarditis (transient, can be lethal)
  - Brain (polio, coxsackie A & B) = paralytic disease, encephalitis → meningitis
  - Eye (enterovirus 70/71) → hemorrhagic conjunctivitis

**Poliovirus** \(\rightarrow\) 90% asymptomatic; 9.9% mild respiratory illness, 0.1% paralysis
- Sx’s of paralytic polio = myalgia → flaccid paralysis (asymmetric, legs more than arms)
  - Most infections in summer and fall
- Virus shuts off host protein synthesis → replicates in motor neurons, causing paralysis
- Fecal-oral transmission (human waste contaminates in water supply)
- 2 types of polio vaccine:
  - Inactivated (currently used in US)
  - Live, attenuated (generates mucosal immunity, but rare revertants to wild-type)
- Thought to be eradicated, but small # cases found in Africa/endemic areas recently

**Togaviruses** \(\rightarrow\) enveloped plus-strand RNA viruses; 2 types:
1. **Alphaviruses** \(\rightarrow\) cause encephalitis (WEE, EEE) or rash and arthritis (Ross River virus)
   - Spread by mosquitoes (geographically restricted distribution)
2. **Rubella** (aka German measles) \(\rightarrow\) causes mild rash illness in adults and children
   - Sx’s → mostly asymptomatic; low-grade fever, lymphadenopathy, conjunctivitis followed by rash on face that spread to trunk and limbs (fades after 3 days)
   - Infection in pregnant women can have severe consequences
     - Fetal infection can lead to “congenital rubella syndrome” (MR, growth defects, heart defects, cataracts, deafness, liver/spleen/bone marrow problems)
     - >99% women can have exposure, viremia, placental infection and/or virus entry into baby’s blood and still have healthy baby
       - Biggest impact in early pregnancy
   - Spread via respiratory transmission, world-wide distribution
**Flavaviruses** → Mosquito-borne viruses (yellow fever, dengue, Japanese encephalitis, West nile)
- Can also be tick-borne; or person-to-person through blood (only Hep C)
- West Nile → mostly infects birds, humans are incidental hosts; no effective treatment
  - Can be asymptomatic, but can cause flaccid paralysis or encephalitis in some patients

**Human Coronaviruses** → Cause the common cold (in addition to rhino- and adenoviruses)
- Can cause SARS (severe acute respiratory syndrome) in the lower respiratory tract (PNA)

### Negative Strand RNA Viruses

<table>
<thead>
<tr>
<th>Positive Strand RNA</th>
<th>Negative Strand RNA</th>
</tr>
</thead>
<tbody>
<tr>
<td>- mRNA-sense → 1st step is translation is using host ribosomes in cytoplasm to replicate virus</td>
<td>Opposite-sense of message → 1st step is transcription to synthesize + strand RNA (mRNA)</td>
</tr>
<tr>
<td>- Use host machinery, don’t bring in own</td>
<td>- Can then use host ribosomes to translate</td>
</tr>
<tr>
<td>- Genome not recognized by host machinery</td>
<td>- Polymerase is part of incoming virion (structural protein)</td>
</tr>
</tbody>
</table>

**Key Features of Negative Strand RNA Viruses**
- Virion contains RNA-dependent RNA polymerases (*genomic RNA is not infectious* because host cell ribosomes cannot recognize it)
- Genomic RNA is packaged in protein; structure = nucleocapsid
- Virions are enveloped (emerge from cell taking some of host membrane with it)
  - Entry into cell by virion fusion or by cell-cell fusion (infected + uninfected cell = syncytia)

**Diseases caused by negative/ambisense (read in both directions) RNA viruses:**
- Wide range of infections/diseases that vary in prevalence and have few preventable vaccines

**Influenza Viruses:** 3 types A, B, and C
- A and B are antigenically distinct, but structurally similar → **A is more prevalent**
  - Both cause disease in adults and children
  - Influenza A can infect ducks, chickens, horses, swine (birds = largest reservoir)

**Influenza virion**
- Outer envelope → derived from host plasma membrane
  - Contains viral glycoproteins **hemagglutinin** (HA) and **neuraminidase** (NA)
- Inner scaffolding → matrix protein, required for virion assembly
- Genome → segmented
  - 8 different strands, each containing 1-2 genes
  - When 2 different flu viruses infect 1 cell, segment reassortment can occur to **produce a new virus** (1 of 2 mechanism of flu genetic diversity) → $8^8$ possibilities

**Influenza gene reassortment cause** **antigenic shift** → pandemic strains can arise from avian reservoirs
- Birds can infect pigs/non-human species → pigs can transmit to humans
  - Rare transmission between birds and humans because avian flu doesn’t work well with human respiratory cell machinery
- Bird and human strain infection in pig ("mixing vessel") → re-assortments can lead to flu with external bird antigens and internal human replication machinery
  - "New" virus can replicate in humans, who lack immunity to bird antigens (many hosts)
- Avian flu 2009 → multiple sources of bird flu in pigs became infectious in humans

**Antigenic Shift** = Large change; replacement of entire hemagglutinin → **pandemic**  
**Antigenic Drift** = Smaller change; single site mutations to hemagglutinin binding site → **seasonal epidemics**

**Hemagglutinin (HA)**
- 17 different antigenic types (Flu A, H1-17) → H1 & H3 infect humans  
  - Multifunctional → binds cellular receptor sialic AND mediates fusion of viral envelope with host cell membrane  
  - Is the target of neutralizing antibodies → minor mutations can result in antigenic "drift"  
  - Replacement with gene from alternate hosts results in antigenic shift  
  - Is synthesized in "pro-form" (cleavage required for conformation change/activation of fusion)  
  - Cleaved by trypase Clara (serine protease secreted by non-ciliated Clara cells)  
    - Clara cells found in bronchial and bronchiolar epithelium in respiratory tract lumen  
    - Possible explanation for restriction of replication to respiratory tract

**Influenza Entry into Host Cell:**  
Virus binds sialic acid (host receptor) → Endocytosed → HA conformational change 2° acidic pH → Exposes hydrophobic domain → Fusion of viral glycoprotein with endosomal membrane → Viral genome pushed out of endosome into host cytoplasm  
At neutral pH (extracellularly), hemagglutinin does not expose the hydrophobic domain → does not fuse with membrane → no syncytia are seen

**Neuraminidase** = Tetramer that cleaves sialic acid residues on cell surface allowing virus to leave
- N1-10 currently infect humans  
- Mutated NA active site → aggregation of viruses on same infected cell (can't spread infection)

**M2 Protein** → Spans the viral envelope and acts as a proton pump  
- Activated while virion is in the acidic endosomal compartment, pumps H+ into virion  
  - Acidification of virion loosens protein-protein contact, facilitating viral uncoating  
- Target for amantadine antiviral no longer used due to resistance

**Influenza Nomenclature** = Type/# of isolate/year of first isolation/HA & NA subtypes  
- Example = A/Hong Kong/156/97 (H5N1)  
- Historically, there was only 1 circulating strain until accidental release of H1N1 lab strain  
  - H1N1 and H3N2 circulating in population

**Influenza Replication** → occurs in ciliated columnar epithelial cells in respiratory tract  
- Large numbers of virions are shed into respiratory tract → facilitates transmission  
- Infection damages the respiratory tract → disrupts mucus layer, epithelium, creation of transudates and exudates (inflammatory and dead epithelial cells)

**Uncomplicated Flu Symptoms** → Fever, cough, rhinorrhea, fatigue, HA, chills, myalgias  
**Complicated Flu** → Flu followed by PNA infection
- Primary viral PNA → worsening of classic sx's, no response to abx, high mortality  
- Secondary bacterial PNA → sx's improve then worsen, response to abx, low mortality
Diagnostics → amplification methods (rapid culture and PCR)

Immunity responses to influenza:
- Innate → mucus barrier, clearance by cilia, alveolar macrophages (impairment of these ↑ risk)
- Adaptive → IgA, IgG protection; clearance by IgG + complement or by CD8 cytotoxic cells

Vaccines → Reformulated annually, provides partial protection (↓ severity if infected)
- Killed vaccine → HA and N antigens; given IM, used in IC patients, and ages 6 months to >69 years
- Live attenuated → replicated restricted to nasopharynx (cold-adapted, temperature-sensitive)
  - Used in healthy, non-pregnant people 2-49 years old

Paramyxoviruses
- Do not undergo antigenic change
- No animal reservoir → maintained in the population by continuous person-to-person spread
  - Spread via respiratory route
- Common among all of these viruses → seasonal outbreaks, severe disease in young/elderly, infections recur throughout life* (sx's less severe in adults)
  - Diagnosed → direct Ag visualization, culture, PCR/amplification
- Includes parainfluenza 1-4, respiratory syncytial virus, metapneumovirus, measles, mumps

Respiratory Syncytial Virus (RSV) → outbreaks of respiratory disease in the winter
- Also causes otitis media, bronchitis, bronchiolitis, croup, and PNA (more severe in babies)
- Spread via direct contact with respiratory secretions (isolation to prevent nosocomial infections)
- Primary URI can progress in severity with dyspnea, increased RR, and hypoxemia (1% infant mortality)

Human Metapneumovirus → causes 7-40% of pediatric respiratory infections, similar to RSV

Parainfluenza Types 1-3 → common cause of URI, most common cause of croup in young children

Measles Virus
- Distributed worldwide; incidence depends on vaccination rates (epidemics can occur in countries with high vaccine coverage; can become endemic in countries w/o good coverage)
- Transmitted via respiratory/aerosol → attack rate is 99.9% (% infected if susceptible)
  - Significant in developing countries → 30% infant mortality rate
- Pathogenesis → Local infection in respiratory epithelium spreads to lymph nodes, replicates and infects monocytes, which disseminate (low level viremia) and infect epithelium throughout body (causing huge secondary viremia and measles symptoms)
- Sx's → fever, cough, coryza, conjunctivitis, rash (due to CD8 cell immune response)
- Diagnosis → Clinical presentation, serology (IgM)
- Prevention = live attenuated viruses given to 12-15 month old infants
  - Has changed age distribution of infection to very young infants and older children - previously seen in children around 5

Mumps → infection of glandular epithelial cells; prevented by live attenuated virus
- Sx's → parotitis, orchitis most recognized; pancreatitis and ovarian infection are less frequently recognized; meningitis in 10% cases
- Diagnose via culture or serology
Rhabdoviruses → Large family that can infect vertebrates, invertebrates, and plants → rabies is only important human pathogen
- Incidence is a function of control in domestic animals → endemic in wildlife (bats, raccoons, etc.)
  - <10 cases/year → mostly important or contact with rabid bats
- Transmitted from saliva in bite of infected animal → limited replication in muscle, but neurons uptake and over 14-90 days, transported to CNS
- Sx’s = progressive neurological disease
  - Prodrome (fever, malaise, paresthesias) → anxiety, aggression, seizures, hypertonia, paralysis → coma and death
- Diagnosis → Clinical history, biopsy/immunochemical staining, PCR
- Prevention → Inactivated virus given post-exposure, due to long incubation period
  - Pre-exposure vaccination for jobs with high risk of exposure (working with animals)

Viral Gastroenteritis

<table>
<thead>
<tr>
<th>Viruses that replicate in the GI tract and DO NOT cause GI disease</th>
<th>Viruses that replicate in the GI tract and cause gastroenteritis</th>
</tr>
</thead>
<tbody>
<tr>
<td>- Enteroviruses (Poliovirus, coxsackie, ECHO, Hep A, Enteroviruses 68-71)</td>
<td>- Adenovirus</td>
</tr>
<tr>
<td>- Reoviruses</td>
<td>- Calicivirus (Norovirus) →+ strand RNA, nonenveloped</td>
</tr>
<tr>
<td>- Adenoviruses</td>
<td>- Astrovirus</td>
</tr>
<tr>
<td></td>
<td>- <strong>Rotavirus</strong> → segmented dsRNA, nonenveloped</td>
</tr>
</tbody>
</table>

Infection in the GI tract is facilitated by viral resistant to low pH (stomach), detergents (bile), and proteases (small intestine)
- Different viruses that cause gastroenteritis have various sites of replication (villi, crypts, M cells)

**Rotavirus** = most common cause of severe dehydrating diarrhea in young children (spike in winter months)

Rotavirus (part of the Reovirus family)
- Genome → 11 segments of ds RNA each encoding 1 protein (RNA alone isn’t infectious)
  - RNA segments form different viruses and may re-assort during dual infections
  - Rotovirus can be typed by the sizes of the different RNA segments
- Capsid Structure → VP4 and VP7 cover most of surface → required for attachment/entry
  - Are also neutralization epitopes (antibodies to VP4/7 can neutralize virus → immunity)
  - Note: VP = viral protein; NSP = non-structural protein

Replication cycle - Rotavirus/double-strand RNA viruses:
Attachment → endophagocytosis → virion leaves lysosome & uncoats → RNA translation OR transcription
- Translated RNA forms virus-specific structural and non-structural proteins
- Transcribed RNA is replicated and packaged into new virions → then LYSIS of cell

Pathogenesis → Infects mature absorptive enterocytes in the small intestine
- Enterotoxin **NSP4 peptide** causes Cl- secretion by secretory crypt cells → watery diarrhea
- After 2-3 days, have villus atrophy/inflammation (lose absorptive function, causing diarrhea)

Diagnosis of viral gastroenteritis → antigen-specific enzyme immunoassay (not easily cultured)
- Usually just diagnosed clinically w/o tests → treated with supportive therapy, no abx

Impact → #1 most important cause of severe dehydrating diarrhea illness in infants/children worldwide
- US: 1 million cases, 150 deaths; developing countries: 150 million cases, 900,000 deaths annually
- Prevention → Currently 2 live, oral, attenuated vaccines (effective, given w/ scheduled vaccines)
  - Prior re-assortment rotavirus vaccine (Rotashield) taken off market 2° intussusception
**Norovirus** → genus that is part of family of Caliciviruses

- Structure = cup-shaped indentations on surface that aid in attachment to host cell

Replication Cycle: Endophagocytosis → endosomal membrane dissolves → viral uncoating → viral RNA is translated and transcribed/replicated → packing and release

Sx's peak within 24 hours of infection → fever, vomiting, rapid transmission = Norovirus hallmarks

- "Secretors" and "non-secretors" have different susceptibilities → non-secretors are protected

**Epidemiology** → Recent outbreaks, particularly in cruise ships

- Nursing homes and hospitals most likely for outbreaks → Significant hospitalizations and deaths in the elderly due to norovirus

- Only ~1000 viruses necessary for infection

<table>
<thead>
<tr>
<th>Rotavirus</th>
<th>Norovirus/Calicivirus</th>
</tr>
</thead>
<tbody>
<tr>
<td>- dsRNA virus that synthesizes RNA inside a transcriptionally active particle</td>
<td>- Plus strand ssRNA virus with genome similar to picornaviruses</td>
</tr>
<tr>
<td>- Most important cause of dehydrating diarrhea in children &lt;2 years old worldwide</td>
<td>- Causes outbreaks of gastroenteritis in all ages</td>
</tr>
<tr>
<td></td>
<td>- Most common cause of infectious GI illness</td>
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<td></td>
<td>- Cruise ships and nursing homes</td>
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</tbody>
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**Viral Hepatitis**

Clinical syndrome of hepatitis → inflammation in the liver

- Caused by toxins, medication, 5 viral infections (Hepatitis A-E) & other infections

- Features of acute hepatitis → jaundice

Typical course of hepatitis:

Exposure → Incubation → Symptoms and jaundice → Recovery or persistent liver infection

- Acute HBV infection with recovery

- Progression to chronic HBV infection

<table>
<thead>
<tr>
<th></th>
<th>Hep A/E</th>
<th>Hep B/D</th>
<th>HEP C</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Transmission</strong></td>
<td>Fecal-oral</td>
<td>Sexual, blood, perinatal</td>
<td>Blood</td>
</tr>
<tr>
<td><strong>Incubation</strong></td>
<td>2-6</td>
<td>6-24</td>
<td>6-300</td>
</tr>
<tr>
<td><strong>% who get jaundice</strong></td>
<td>30-70</td>
<td>20-40</td>
<td>15-25</td>
</tr>
<tr>
<td><strong>% persistent infection</strong></td>
<td>0</td>
<td>5</td>
<td>70*</td>
</tr>
</tbody>
</table>

Differentiating between hepatitis viruses

1. A & E → lack lipid envelope; not disinfect by bile (fecal-oral transmission); self-limiting disease

2. B & D → Sexual, blood, and perinatal (all except fecal-oral); persist only in patients who get it perinatally because immune system tolerates virus

3. C → Blood exposures (needle sharing, medical procedures); 70% get persistent infection

   a. Chronic infection → Fibrosis that bridges portal triads, forming nodules and cirrhosis

   b. Also increased rate of CA; either way, liver will fail, pt will die w/o transplant

Diagnostic tests and indication → ALT/AST (liver inflammation), IgM (acute infection), IgG (chronic infection), viral particles/ protein & nucleic acid (ongoing infection)
**Hepatitis A Virus** → most common cause of acute hepatitis in US
- Capsid proteins elicit universal neutralizing antibody (1 serotype)
- Diagnosed via IgM anti-HAV; prevention via vaccination or prior infection

**Hepatitis B Virus** →
- Genome → Circular, dsDNA; can integrate into host genome like a plasmid
  - S gene contains neutralizing epitopes that prevent infection → Used to make 1st recombinant and 1st cancer vaccines
    - Liver CA in children ↓ by HBV vaccination
- HBV genome hides in the nucleus of cells → difficult to find eradicate (infection reservoir)
  - HBV drug treatment → must kill new microbes AND ones hiding in cell nuclei
  - Circular DNA still in hepatocyte sustaining infection → may return if abx are stopped
- Virus is concentrated in blood and stable in the environment (highly transmissible)

Hepatitis B surface antigen can be used to protect against cancer
- Recombinant HBsAg vaccine is safe and effective at preventing cancer
  - Decreased incidence of liver cancer in children in Taiwan with HBV vaccination
- Lipid envelope and glycoprotein spikes with a neutralizing determinant

**Hepatitis D Virus** → RNA genome, fully dependent on outer coat from Hep B (HBsAg) to complete lifecycle
- Only seen clinically with acute or chronic Hepatitis B co-infection

**Hepatitis C** → the most diverse genome that infects humans (moreso than HIV)
- **Host genetic diversity** determines infection outcome
  - Viral recovery can occur, eliminating all long-term complications
  - Frequency of IL28B allele explains worldwide differences in spontaneous clearance of HCV
    - **IL28B** = Single polymorphism that confers protection against Hepatitis infection
- **Viral genetic diversity** allows viral persistence and evasion of treatment/eradication
  - Clinical challenges = 70% HCV persistence, treatment resistance, and vaccine development
  - Replication → rapid turnover with error-prone replication, allow development of variants that can evade immune response or treatment

**Hepatitis E** → Acute infection has a high fatality rate in pregnant women; doesn’t cause chronic hepatitis
- In healthy adults, is normally a self-limiting illness

**Retroviruses**

HIV-1 was discovered in 1983 → an RNA containing virus with reverse transcriptase activity

Patients with AIDS have decreased CD4+ T cells, increasing the % of CD8+ T cells
- Opportunistic infections occur when CD4 < 200 (an AIDS-defining condition); normal CD4 ~1000
- Dramatic drop of CD4 cells in weeks 1 & 2 → CD4 count is the #1 most important clinical measure
  - Level of initial CD4 damage and rebound predicts progression of disease
- Viral load in plasma is second most important clinical measure
  - **Viral set-point** (lowest level after acute infection) predicts disease progression → higher set point = better prognosis

Blood tests for HIV → Detection of host antibodies via SDS-PAGE & Western Blot, ELISA
- Essential for testing blood donors to prevent spread through transfusion
- Quantitative RT-PCR is most accurate, used today in addition to ELISA
Retrovirus Phylogeny

- Complex retroviruses encode regulatory genes as well as \textit{gag}, \textit{pol}, and \textit{env}
- HIV-1 is a \textbf{lentivirus} vary greatly within a host
  - No lentiviruses are controlled by neutralizing antibody response due to antigen diversity
- Origin \rightarrow HIV-1 from chimpanzee; HIV-2 from mangabays \rightarrow cross-species transmission led to human epidemic
  - HIV is most closely related to SIV (simian immunodeficiency virus)
  - SIV does not cause disease in its natural hosts (but does in other monkey species)

Essential Elements of Retroviruses

- Enveloped, with RNA genome & viral reverse transcriptase (RT) and integrates into host genome
- 3 major genes
  - \textit{gag} \rightarrow structural proteins, make up the core of the lentivirus
  - \textit{pol} \rightarrow enzymes (protease, RT, integrase)
  - \textit{env} \rightarrow coat protein (2 proteins gp120 and gp41)
- Complex retroviruses like HIV also encode accessory genes (\textit{Tat}, \textit{Rev}, \textit{Nef}, \textit{Vif}, and \textit{VPR})

HIV Virion

- Enveloped \rightarrow membrane is derived from host cell, contains viral and host proteins
- Gp120 (glycosylated protein) mediates attachment \rightarrow is a target for neutralizing antibodies
- Virus contains 2 copies of RNA and all proteins required to transcribe to DNA and integrate into host genome
- Gag protein is the most abundant protein in the virus \rightarrow processed to create structural proteins and protease (Gag can be used for virus detection)

Medications can target this process or ANY step in the HIV life cycle

HIV Life Cycle:
1. Attachment/Fusion (requires receptor and co-receptor)
2. Reverse transcription of RNA to cDNA
3. Integration of viral DNA into host genome
4. Viral gene expression
5. Assembly and budding
6. Maturation

1. In addition to CD4, a \textbf{co-receptor} is required for entry into host cell, and determines virus tropism
   - Interaction with \textbf{CD4} on target cell induces conformational change in \textit{gp120}
     - This causes a conformation change in transmembrane protein \textit{gp41}, which mediates fusion with target cell
   - Co receptor = chemokine receptor \textbf{CCR5} \rightarrow seen in early infection, on T cells and macrophages
   - Virus eventually mutates to also recognize \textbf{CXCR4} (only in T cells) later in infection
     - CXCR4 utilization makes virus more T-cell tropic and is a \textbf{bad prognostic indicator} (more efficient T cell infection and rapid drop in CD4 count)

2. Reverse Transcription of Viral RNA to DNA
   - At 5’ end there is a primer binding site, where a tRNA binds and together with the RT enzyme, reverse transcription in virion begins
     - There is a repeated R region at 5’ and 3’ ends \rightarrow complementary DNA is made, and binds to the identical sequence RNA
• Need 2 RNAs because while one is being transcribed, one binds to new, complementary DNA
  o RNAase H degrades RNA associated with DNA, leaving only DNA behind
• End up with double-stranded DNA with 3 copies of U5 and U3 region (long-terminal repeats → facilitate transcription of complementary RNA)

3. HIV integration into host genome:
• **Viral integrase** in virion acts an exonuclease (cuts 2 bases at end of viral DNA) and endonuclease (opens host genome and enters at random)
  o Repair is done by host cell proteins
• Clinical significance → Once integrated, HIV remains in host cell until it dies

4/5. Protein Translation and HIV Assembly
• HIV viral protein transcription by host cell depends on active (open chromatin) versus latent (condensed chromatin) state of host cell
  o Virus can survive in latent cells, host will not see it as foreign b/c no proteins are made
• In active cells, RNA is transported to cytoplasm and is translated by host ribosomes
  o Envelope proteins are processed in ER and golgi, inserted into cell membrane

6. HIV Virus Maturation → Essential in producing infectious virus
• **HIV protease** allows maturation of newly released virions to an infectious form
  o Protease cleaves itself from Gag-pol precursor, then cleaves precursor proteins liberating virion structural proteins and enzymes
• Specific protease inhibitors are important in HAART

Must treat with multiple antiviral drugs given the high rate of mutation in HIV/all retroviruses
• High rates of mutation, and reverse transcriptase has NO proofreading/editing function
• Virus resistance arises with partial viral suppression
  o Partial suppression allows for some replication (mutation) to continue, and selective pressure to provide resistant mutants an advantage
• Statistics → unlikely to develop resistance to 3 different drugs in the same viral genome

Transmission of HIV → Probability of transmission depends on viral load in direct inoculation
• Virus particles are fragile and therefore require direct inoculation
  o **Sexual** transmission
  o **Contaminated needles** (IV drug use, needle stick injuries)
  o **Mother-to-child** transmission (in utero, at delivery, breastfeeding)
• Is not transmitted by aerosols or skin contact

Human Genetic Variation Alters Response to HIV
• Prevent viral entry into cells → CCR5Δ32 prevent infection (no CCR5)
  o Alternatively, CCR5 P1 increases CCR5, increases infection and viral load
• Improved innate immune control slows disease progression → KIR + HLA B*57 and B*27
• Improved adaptive immune control → HLA B*57 and HLA B*27

HIV latency in memory T cells establishes a long-lived infection that isn’t cleared by HAART

The Berlin patient → HIV+ who developed lymphoma; received bone marrow with homozygous Δ32CCR5 mutation (no CCR5 expressed), was taken off HAART and effectively cured (no detectable HIV RNA/DNA)
• 2nd case in infant that was cured with early treatment at birth
HIV Epidemiology and Clinical Aspects

Epidemiology
- In 2011, 35 million infected and 1.7 million deaths from HIV (most in Sub-Saharan Africa)
- Incidence (# new cases each year) in US remains constant (50,000 each year)
  - Main transmission via MSM contact, #2 is heterosexual contact (though in Baltimore ~50% due to drug use)
  - % diagnoses and new infections higher in African Americans, mostly men
- Likely many contributors to the race/ethnicity disparities in HIV prevalence

Transmission
- HIV is highest in genital secretions, blood, and breast milk
- Sexual transmission
  - Risk highest with acute infection (high viral load), lowest with undetectable viral load
  - Decreased risk with condom use, type of sexual activity, circumcision
  - Increased risk with concomitant STD infection
- Blood transmission → IDU, PEP (occupational exposure), transfusions
  - W/o treatment, 15-30% risk to pass to baby; w/ treatment, 5-20% chance
- Mother-to-child → can occur any time during pregnancy, delivery, breastfeeding
  - Virtually eliminated in high-income countries, but is majority transmission in low-income countries

Prevention
- Vaccines ⇒ much research, no success
  - Use of adenovirus 5 as a vector ⇒ pre-existing antibodies increased risk of HIV
  - Thailand study ⇒ Some efficacy, but not enough to stop epidemic
- Pre-exposure prophylaxis with ART (PrEp) ⇒ Oral and topical ART
  - Treating high-risk population to try to prevent infection; mixed results
  - Reduced incidence related to drug adherence, most participants weren’t adherent
- Post-exposure prophylaxis (PEP) ⇒ 0.3% chance of transmission with needle-stick injury from HIV-positive patient; most effective if started immediately, <1 hour
- Male circumcision ⇒ ~50% decreased risk of HIV infection in circumcised men
- Treatment as Prevention ⇒ Chance of transmission to partners is lower with low viral load
  - Treatment of HIV+ patients = 96% decrease in transmission
- Hypothetically with universal voluntary HIV testing and immediate ART ⇒ little/none HIV
  - Treatment cascade ⇒ Small % of patients who have HIV, have been diagnosed, have access to care and are adherent to care

Stages of HIV Infection:
1. Primary Infection
2. Acute HIV syndrome ⇒ 1st few weeks, wide dissemination; seeding of lymphoid organs
3. Clinical Latency ⇒ average 8 years, virus decreases to viral load set point
4. Constitutional symptoms
5. Opportunistic diseases ⇒ Onset of AIDS until death ~1.5 years

Diagnosis:
- Acute infection ⇒ HIV RNA PCR (“viral load”)
- Acute to subacute infection ⇒ p-24 antigen assay
- Subacute to chronic infection ⇒ ELISA followed by Western Blot to confirm
**Acute retroviral syndrome** arises during initial acute infection (high viral load) with flu-like symptoms and goes away during seroconversion (mounted antibody response):
- Sx's vary widely → almost all have fever, most have adenopathy, pharyngitis, rash, and myalgias
- Less common = HA, N/V/D, hepatosplenomegaly, weight loss, thrush, neurologic symptoms

**Clinical Evaluation and Treatment**

AIDS = HIV Infection plus ONE of the following:
- 1 of 23 AIDS-defining illnesses (16 infections, 5 CAs, wasting, encephalopathy)
- CD4 count < 200
- CD4 count < 14% total lymphocytes

**CD4 count** provides a real-time estimate of a patient’s risk of developing complications

**Viral load set point** related to how quickly disease will progress (high set point = faster progression)
- Viral load set point not used much clinically anymore, pts started on HAART immediately

Opportunistic infections that are common in AIDS patients = cryptococcal meningitis, *Pneumocystis carinii* pneumonia (PCP), *Mycobacterium avium* complex (MAC)
- At CD4 ≤ 200 → prophylaxis treatment against PCP (bactrim) → *know these #s/infections*
- At CD4 of ≤ 50 → prophylactic azithromycin against MAC infection

**HAART** = highly active antiretroviral therapy (or ART = antiretroviral therapy)
- Became standard of care in 1996
- Current recommendation to start in all patients with CD4 ≤ 500, some Drs offer to all HIV+ pts
  - More evidence that untreated HIV has negative consequences at all stages of disease
    - Increased MIs, strokes, cognitive decline, renal failure
  - New drugs and combinations are better tolerated, have better efficacy and adherence
- HIV+ 20-year old have life expectancy of 51 (general in US for men = 57)
  - Lowest life expectancy in drug users with HIV because of co-morbidities

**Treatment Goals**
- Decrease the viral load in serum to undetectable (currently < 20)
- Preserve immune function (prevent deterioration)
- Avoid morbidity and mortality due to HIV/AIDS
- Minimize medication side effects
- Decrease transmission/spread

**Anti-HIV Drugs**

Know for the exam:
- MOA for nucleoside analog reverse transcriptase inhibitors (NRTI’s), non-nucleoside reverse transcriptase inhibitors (NNRTI’s), and HIV protease inhibitors (PI’s)
- Know the following specific drugs in detail (including pharmacokinetics and toxicities):
  - Zidovudine (AZT, azidothymidine)
  - Nevirapine
  - Ritonavir
Nucleoside Analog Reverse Transcriptase Inhibitors (NRTIs) → 8 approved including Zidovudine/AZT
• Initially discovered from cancer treatment research; all have similar structure (like nucleosides)

Mechanism of Action
• Must be phosphorylated by cellular enzymes to triphosphate (AZT-TP) in order to work
• AZT-TP inhibits HIV-encoded reverse transcriptase (RNA-dependent DNA polymerase)
• Is structurally related to DNA (thymine) → acts as a chain terminator when incorporated
• Reduces plasma HIV-1 RNA by 0.5 log in vivo by itself, but still effective

Pharmacokinetics
• Well-absorbed; Eliminated via glucordonidation (Phase II enzyme conjugation)
• Short plasma half-life of parent compound (1 hr), longer half-life of intracellular AZT-TP (4 hrs)
• Rapid conversion to the monophosphate (AZT-MP) which accumulates in the cell; slow conversion to di- and triphosphates
  ○ Can have less frequent dosing because plasma level isn't what matters

Toxicity = mainly bone marrow suppression → mainly anemia, but also less commonly granulocytopenia
• Rarely causes myopathy and lactic acidosis/steatosis (potentially fatal)
  ○ Mitochondrial toxicity due to poor selective toxicity → inhibits mitochondrial DNA polymerase more effectively than nuclear DNA polymerases

Resistance is slow to occur (months/years) → requires 5 or more specific amino acid changes
• There is limited cross-resistant with other nucleosides
  ○ Can use other NRTIs if AZT fails (e.g. Tenofovir/PMPA → most common HIV drug in world)

Non-Nucleoside Reverse Transcriptase Inhibitors (NNRTI’s) → 5 approved including Nevirapine
• Specifically designed to treat HIV, unlike NRTI’s, all have different structures

Mechanism of Action
• Already active, don’t require intracellular activation by enzymes like NRTIs
• Act as non-competitive inhibitors → binds to a site that isn’t the active site and induces conformation change in reverse transcriptase enzyme, rendering it inactive
  ○ Does not bind same site in HIV-2 RT → can’t treat HIV-2, only HIV-1
  ○ Reduces plasma HIV-1 RNA by 2-3 logs (short-lived effects as monotherapy)

Pharmacokinetics
• Well-absorbed; eliminated by oxidative metabolism (cytochrome P450 3A4)
  ○ Is a P450 enzyme inducer → possible drug-drug interactions
• Long plasma half-life (25-30 hrs), allowing infrequent dosing (bid or qd)

Toxicity → not very toxic due to good selective toxicity (no effect on human DNA polymerase)
• Hypersensitivity rash is common, but is usually transient
• Rarely can cause Stevens-Johnson syndrome (lethal skin-sloughing syndrome), hepatotoxicity (more common in pregnant women), and drug interactions (p450 induction)

Resistance occurs easily and emerges rapidly (days/weeks)
• 1 specific AA change can confer up to 1000-fold resistance to the drug
• There is cross-resistance to other NNRTI’s

In the US, use Efavirenz instead of Nevirapine → better tolerated, longer half-life, less toxicity, side effects are CNS/psych symptoms (dreams, psychosis, etc.)
• Was part of the first 1-pill a day regimen for HIV treatment
HIV Protease Inhibitors → 9 on the market, Ritonavir

Mechanism of Action
- Already active, like NNRTI's → doesn't require intracellular activation
- Act as competitive inhibitors of HIV proteases by essentially freezing the enzyme in the high-energy transition state → prevent activation/cleavage of HIV proteins
  - Prevents maturation of HIV virion → are still released by cell, but aren't infectious
- Reduce plasma HIV-1 RNA by 2-3 logs as monotherapy
- Partially restore CD4 cell count (average increase 100-150) even as monotherapy (when used with other drugs can restore CD4 count to normal)

Pharmacokinetics
- Variable bioavailability (40-60%) due to first-pass metabolism and autoinduction (induces its own metabolism) → highly protein bound (99%)
- Eliminated by oxidative metabolism (Cytochrome P450 3A4) → potent P450 enzyme inhibitor and hepatic enzyme inducer
- Plasma half-life is 3-4 hours, dosed BID

Toxicities → good selective toxicity, do not inhibit human aspartyl proteases
- GI intolerance (N/V/D), hyperlipidemia, glucose intolerance (rarely)
- Associated with fat redistribution
- Paresthesias in fingers and lips common during first few weeks
- Drug interactions due to P450 enzyme inhibition
  - Can be advantageous → dramatically reduce clearance of other HIV drugs, reducing the frequency of dosing of other meds

Resistance → intermediate between NTRIs and NNTRIs (weeks/months, but not in all pts)
- 1 AA change confers a 3 to 5-fold resistance to the drug (primary resistance mutation)
- Secondary mutations accumulate and confer increasing resistance, up to 100-fold
  - Accumulated mutations can confer partial or complete cross-resistance to other PIs
- Dose-response → higher doses of drug suppress emergence of resistant phenotype

Other antiretroviral drugs → (not for exam):

Integrase Inhibitors → Raltegravir
- MOA → inhibits HIV integrase/DNA strand transfer reaction necessary for integration
- Given PO BID, highly potent and well-tolerated with little/no toxicity

Entry Inhibitor → Enfuvirtide
- MOA → Interferes with protein helix bundle formation necessary for membrane fusion/entry
- Given IM BID, very expensive, only used as last treatment

Entry Inhibitor → Maraviroc
- MOA → Selectively inhibits HIV entry mediated by CCR5 chemokine receptor
- Given PO BID, only effective in patients with CCR5-tropic HIV

HAART Today
- Combination therapy only → strictly to prevent drug resistance of HIV
- Current trends = reduced bill burden, once-daily regimens, long-term tolerability
Concentration dependent = fluoroquinolones and aminoglycosides
PAE → all β-lactams for gram +; carbapenems for + and -, aminoglycosides for + when used synergistically

Know MOA, toxicities/side effects, main resistance mechanisms (efflux, mutation, etc.), spectrum of activity for each drug

Specifically know the drugs that cover S. aureus/MRSA and Pseudomonas

Beta-lactams → ALL are cell wall agents (vancomycin included), time-dependent, PAE for gram + (except carbapenems also for gram -)

Penicillins
PCN G → limited activity, poor bioavailability
Aminopenicillin → Increase bioavailability, slightly increased coverage of gram +/-(Ampicillin)
Antistaphylococcal PCN → activity against Staph that produce penillinase, but not ones that have mutations in PBP that lead to MRSA resistance (oxacillin, methicillin)
Antipseudomonal PCN → increased coverage to psuedomonas (piperacillin)

*Adding a beta-lactamase inhibitor broadens coverage and adds anaerobic coverage as well as bacteria that produce beta-lactamase
  • Zosyn (piperacillin/tazobactam) → gram +/-, pseudomonas, anaerobes (ALL except MRSA)
  o Negative consequences for microbiome
**PCNs can be used to treat all Strep, iffy for S. pneumoniae

Cephalosporins → time dependent, cell wall active agents, PAE gram positive mostly
  • 1st generation → gram positives, Strep and MSSA; NOT MRSA
  • 2nd generation → gram positives, anaerobes, some gram negatives (only one to cover anaerobes)
  • 3rd generation → gram positives and negatives
  • 4th generation → gram positives, negatives, pseudomonas
  • 5th generation → gram positives including MRSA, gram negatives
*Cephalosporins don’t cover enterococci or listeria
**Increasing gram negative coverage through the generations

Carbapenem → VERY broad activity; gram positives (NOT MRSA or enterococci), gram negative, anaerobes
  • Better coverage than Zosyn against resistance gram-negatives (best to treat these)

ALL ABOVE ABX ARE BETA-LACTAMS (PCN, cephalosporins, carbapenems)

Monobactram (aztreoname) → cell wall acting time-dependent agents, treats gram negatives and pseudomonas (NOT gram positive or anaerobes), no significant PAE
  • Used in patients with PCN allergy for gram negative aerobic infections (no cross reactivity)

Vancomycin → Inhibits cell wall synthesis; treats MRSA (IV) and C. diff (PO)

Daptomycin → Know that it is DIFFERENT because it disrupts cell membrane; same coverage as Vancomycin → Does not penetrate the lungs
Aminoglycosides ➔ Protein synthesis inhibition, concentration-dependent
- Good PAE against gram positive AND negative, only used with gram positive in COMBINATION with cell wall inhibitors (synergy)
- Significant ototoxicity and nephrotoxicity ➔ given once per day for rapid killing and reduce side effects due to PAE

Tetracyclines ➔ Inhibit protein synthesis, use CA-MRSA and atypical bacteria (chlamydia, borrelia, brucella) ➔ side effect = teeth discoloration in young children

Chloramphenicol ➔ protein synthesis inhibitor, side effect = aplastic anemia, gray baby syndrome
- Antagonism with beta-lactam antibiotics

Clindamycin ➔ protein synthesis inhibition; anaerobes (respiratory tract) and gram positives (some MRSA strains, CA-MRSA), used to treat skin/soft tissue infection; side effects = diarrhea, increases risk of C. diff

Linezolid ➔ protein synthesis inhibition, best gram positive aerobic coverage, including MRSA and Enterococci (worst of the worst); concerning side effect = bone marrow toxicity

Bactrim (TMP/Sulfa) ➔ treats bacteria and fungus (PCP), all CA-MRSA, Nocardia, not good Streptococcal coverage
- HIV+ pts with CD4<200 treated with Bactrim to prevent PCP

Macrolides ➔ protein synthesis inhibition, mainly used to treat pulmonary infections, activity against NTM, some atypical bacteria (chlamydia, mycoplasma, legionella)
- HIV+ pt CD4 < 50 given macrolide to protect against MAC

Fluoroquinolones ➔ MOA gyrase and topoisomerase (level of DNA); concentration dependent; usually benign, but side effects include tendon rupture (unique), CNS effects, especially in elderly pts
- Early generations cover gram negative rods, including some Pseudomonas
- Later generations have broader coverage of gram negative, gram-positive, and anaerobes, atypicals ➔ used to treat respiratory infections

Pseudomonas ➔ 4th generation cephalosporins, antipseudomonal penicillins, carbapenem, monobactam, aminoglycosides

MRSA ➔ 5th gen cephalosporin, Vancomycin, Daptomycin, Clindamycin (for some), Linezolid, Tetracycline, Bactrim (only PO tx for MRSA)

Anaerobes – carbapenems, 2nd gen cephalosporin, beta-lactam/beta-lactamase inhibitor combo
Herpesviruses: Alpha and Beta Herpes Viruses

Common similar features of Herpesvirus → Morphology, ubiquitous, often asymptomatic infection, common modes of replication, all establish latent infections

1. **Alpha** → cause lesions (HSV-1, HSV-2)
2. **Beta** → cause disseminated disease (CMV, HHV6, HHV7)
3. **Gamma** → capable of causing tumors (EBV and HHV8)

Huge genome compared to other viruses (>100kb, >50 genes) → lots of nonstructural proteins
Very common especially in young adults

2 modes of infection:

**Lytic Infection:** Virion entry via fusion with cell membrane → nucleocapsid transport to nucleus → nuclear events (viral gene transcription, genome replication, progeny assembly) → nucleocapsids bud from nucleus (viral envelope forms from host nuclear membrane) → progeny release via exocytosis

Many genes are expressed, occurs in temporal cascades

1. Immediate early → regulate viral gene expression
2. Early → proteins required for genome replication
3. Late → virion structural proteins

**Latent Infection** → no virion production; reservoirs of recurrent disease (recurrence occurs as a consequence of renewed replication or cell proliferation)

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<thead>
<tr>
<th>LYTIC INFECTION</th>
<th>LATENT INFECTION</th>
</tr>
</thead>
<tbody>
<tr>
<td>Many genes expressed in a temporal cascade</td>
<td>Few genes expressed</td>
</tr>
<tr>
<td>Many cell types are infected (2 or more)</td>
<td>Few (1 or 2) cell types infected</td>
</tr>
<tr>
<td>Virion production</td>
<td>No virion production</td>
</tr>
<tr>
<td></td>
<td>Reservoirs of recurrent disease</td>
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Transmission

- **Natural Modes** → Contact between skin/mucus membranes, via secretions (oral, respiratory), OR transplacental
- **Iatrogenic Modes** → transfusion, transplant

Disease severity can vary (higher with 1° infection and IC pts; lower with recurrent infection)

**Herpes Simplex Virus**

- 2 most common presentations = Herpes labialis (oral) and genital herpes
  - Reactivation can be symptomatic or asymptomatic
- HSV-1 and HSV-2 can both occur in genital and oral infections

1° infection in epithelial cells that first come into contact with virus → virions are released and picked up by neurons → infection of neuron cell bodies by retrograde transport → remains in sensory ganglia → event (cold, stress, UV light) activates viral DNA and causes re-infection

Genital Herpes → Incidence = 500,000 cases/yr; prevalence = 40-60 million cases in US
- Correlates with # of sexual partners; women more susceptible than men
- Transmission is commonly unrecognized (asymptomatic shedding is common; 70% cases acquired from asymptomatic partner)
  - Shedding decreases over time with the disease
- Recurrence can occur after years of latency → sx’s usually less severe than in primary disease
• HSV-2 is typically worse and cause genital herpes; HIV-1 is less severe and mainly causes oral herpes → HSV-2 causes more recurrences

Other HSV-2 Presentations → primary gingivostomatitis, herpes whitlow
Can cause HSV Keratitis (# 1 cause of infectious blindness in the world)
• Caused by inoculation into eye OR reactivation through trigeminal nerve

Neonatal Herpes → transmitted through infected birth canal; women asymptomatic in labor
• Causes 3 syndromes: SEM (skin, eyes, mouth), encephalitis, or disseminated symptoms
• Occurrence of vesicles occurs with all of the syndromes, but not always

Herpes Encephalitis → Most common acute encephalitis in US (10-20% of cases)
• Mostly caused by HSV-1
• Classic presentation = fever and focal neuro deficits; usually temporal lobe involvement
  o Temporal lobe enhancement seen on Head CT classically

Varicella-zoster Virus
• Primary infection = Chicken pox
  o Severe disease in newborns/IC patients (PNA, encephalitis, progressive/disseminated dz)
  o Teens and adults (at risk for varicella PNA)
• Reactivation = Herpes zoster (shingles) → lesions localized to innervated dermatome
  o Can disseminate in IC patients (>1 dermatome, cross midline)

Cytomegalovirus → infection usually asymptomatic but disease can occur
• Increased risk in transplant patients, leukemia/lymphoma pts, AIDS pts
• CMV Mononucleosis → 80% mono caused by EBV, 20% by CMV
  o Frequent manifestation of primary infection in young adults
  o Sx’s = fever, lymphadenopathy, lymphocytosis, exudative pharyngitis
• Congenital CMV; severity in baby depends on mother’s infection:
  o If primary infection, severe sx’s seen in 25% babies
  o If reactivated infection, usually asymptomatic in babies (but late onset hearing loss)
• In IC patients:
  o HIV → reactivation and disease with CD4 < 50 (now mostly prevented with HAART)
  o Bone Marrow transplant → commonly PNA after marrow graft
  o Solid organ transplant → large problem, manifests in allograph; highest risk in CMV seronegative recipient and seropositive donor

HHV-6 and HHV-7 → causes rash-like illness in children: roseola infantum (exanthum subitum)
• Can also cause febrile seizures, infectious mono, hepatitis, neurologic sx’s in kids
• May be a problem and cause of a variety of symptoms in immunocompromised patients
• May contribute to disease progression with HIV-1

Herpesvirus Diagnosis → viral culture, antigen detection, nucleic acid detection, antibody detection
• Can culture HSV from all sites except CSF
Human Gamma herpes viruses: EBV and KSHV

Gamma herpes viruses cause disease (esp. cancer) during latent infection, unlike alpha and beta

**EBV Infectious Mononucleosis**
- Transmitted via saliva; 95% adults infected (infection in childhood usually asymptomatic)
  - Infection in adolescence/adulthood associated with syndrome of IM
- Spread via proliferation of infected B cells (virus is latent, not producing virions)
  - Present as circular dsDNA in latent state (made by host cell DNA polymerase)
  - In virion particle, genome is linear dsDNA (requires viral DNA polymerase)
- Sx’s → sore throat, lymphadenopathy, lymphocytosis
- T and NK cells respond to infected B cells; kills many of them
  - In most healthy seropositive pts, >1% of T cells target EBV antigens
  - Many EBV genes are really good T cell targets
    - Only EBV that survives are latent in resting B cells → occasionally will lyse and try to re-infection, but T cell response controls before another infection occurs
- Diagnosis → heterophile/monospot antibody test, IgM to viral capsid antigen, IgG (past infection)
  - Heterophile antibody resolves weeks to months after symptoms
  - IgG detectable for life after initial EBV infection
  - Doesn’t target viral antigens → unknown pathogenesis

**EBV and Immunocompromised Patients → EBV lymphoma**
- Organ transplant recipients are at increased risk for EBV-driven B cell lymphoma due to immunosuppression → if suppression can be stopped, tumor will often resolve
- Congenital immunodeficiency → pt’s often die from EBV lymphoma that develops after EBV infection due to lack of T cell response
- AIDS patients → ~½ lymphoma in AIDS patients is due to EBV
- In contrast X-linked agammaglobulinemia (unable to make B cells) → NO lymphoma b/c NO B cells to infect

**EBV and Cancer → EBV is used clinically to immortalize cell lines**
- Nasopharyngeal carcinomas → always EBV-associated; world-wide
- Hodgkin’s lymphoma → sometimes associated with EBV (30% of cases)
- Post-transplant lymphoma → usually EBV-associated; often in transplanted organ
- Burkitt’s lymphoma → EBV associated in malarial areas of Africa, but not N America/Europe
  - Tumors involving maxilla, periorbital region, mostly in boys; seen in the “malaria belt”
- In tumor cells → EBV DNA/RNA detected; antigen expressed; present at each tumor site at presentation and relapse

**KSHV (Kaposi Sarcoma Herpesvirus)**
- Affects children in Africa, old men in the Mediterranean, organ transplant recipients, AIDS patients
- Tumor typically involves the skin, but can also involve GI tract or lungs
  - Characterized by neovascular proliferation (accumulation of malignant epithelial cells)
  - Purple color from red cells in neovascularure
- Virus discovered by finding herpesvirus-like sequence in tumor cells
  - Carries several genes that closely mimic humans genes (viral IL-6)
- Detected by PCR in B cells of seropositive pt’s (does not immortalize B cells in vitro like EBV)
  - Very few Americans are infected; more prevalent in other parts of the world (S Italy)
- Transmission → Early childhood via saliva, in endemic regions
  - MSM sexual transmission, possible/rarely from IV transfusions or drug use
- KS Pathogenesis → KSHV infection required, most often in IC patients (age, HIV, transplant)
**Primary Effusion Lymphoma** → rare, cancerous B cells floating in pleural or peritoneal fluid (an effusion)
- Occurs in AIDS patients, requires dual infection by EBV and KSHV in the same tumor cell

**Mycobacteria Review: TB, NTM, Nocardia, Actinomyces**

TB, NTM, Nocardia, and Actinomyces are all taxonomically related → a lot of disease overlap (often listed with one another in a differential diagnosis)
- All cause granulomas
  - *M. leprae* and *M. tuberculosis* complex are the only obligate human pathogens

TB is only spread by active infection → whether you are infected depends on how much bacteria you are exposed to and for how long (only ~10% people living with TB+ pts in active infection also get infected)
- Biggest risk factor in US is being foreign-born

ID’ing TB → difficult b/c can present as ANYTHING; but CLASSIC = **Right upper lobe cavitary lesion** on CXR
- In HIV patients, presentation can be atypical and CXR can even be normal!
  - Evaluate for TB in HIV+ pts if **any one** of these → fever, cough, night sweats, weight loss

**TB Workup**
1. **AFB Smear Microscopy** indicates how infectious a patient is (+ test indicates higher bacteria in sputum)
   - Not super specific (40-60%), even less for HIV patients (20-60%)
   - Not specific for TB → could be positive for NTM
2. **Molecular tests/assays** have increased sensitivity, and are fast; but very expensive
3. **Culture** has the highest sensitivity; but SLOW → must make decisions before you have results

**TB Treatment** → **Initial phase** 4 drugs for 2 months; **continuation phase** 2 drugs for 4 months
- Use multiple drugs because each do different things, also prevents resistance
- DOT (Directly observed therapy) → someone provides and watched TB pts take meds every day

**BCG** is the most commonly used vaccine in the world (0-80% efficacy in adults)
- No way to tell if + tuberculin skin test is from vaccination or from latent TB infection → BCG status is ignored when testing for latent TB infection in adults, mostly

**NTM Review**
Slow growers:
- *M. avium* → CD4 <50; pulmonary MAI in older women; cervical lymphadenitis (children)
- *M. kansaii* → looks like pulmonary TB
- *M. marinum* → skin and soft tissue, aquariums

*If pulmonary + CNS symptoms, think **Nocardia**

**Cases:**
1. 44yr old AAF from Baltimore; brother w/ hx of active TB 2 yrs ago) with cough and fever
   - Tests → CXR shows RUL non-cavitary lesion; sputum gram stain → gram positive cocci, negative cultures, negative HIV test, negative PPD test, normal WBC
     - Strep pneumo most likely/CAP → beta-lactams, macrolides, fluoroquinolones
     - Pt started on Moxifloxacin → improves, fever resolves
   - Returns 2 months later with same symptoms
     - CXR → Diffuse infiltrates, PNA on L, cavitary lesion in RUL; NOW workup + for TB
• PPD don’t distinguish latent from active TB; have poor sensitivity (70%) → false negative with active TB
  o DON’T use PPD to r/o TB; must do TB workup (AFB smear microscopy, culture, assay)

2. 25-year old from France with fever → + HIV test with CD4<50; blood culture tested + for TB
• Starts on TB and HIV treatment (not resistance) → patient’s condition worsens
  o **Immune reconstitution syndrome** → HIV meds allowing body to mount an immune response (high fevers, SOB, pulmonary infiltrate)
  o Usually start pt on steroids to slightly immunosuppress transiently

3. 25-yr old US-born medical student; close contact with pt of a smear-positive TB case
• Latent TB test: PPD – positive → but normal CXR, normal liver enzymes
• Positive PPD in high risk situation → treat for latent TB!

4. 32-yr old born in Ecuador; immigrated at age 2, chronic smoker → about to start work in hospital, is asymptomatic but has a + PPD test
• CXR shows small left effusion → Do a TB workup with AFB smear, culture, and molecular tests
• Sputum was negative x3, culture was negative → pleural biopsy was positive for TB

5. 55-yr old female, previously healthy, with SOB and CP; unremarkable PE
• Normal CXR; Sputum AFB smear positive x 1, but negative x2; AFB culture positive for MAI
• Do nothing or repeat tests → DON’T start treatment for MAI because diagnosis requires multiple positive cultures and clinical sx’s (NTMs can contaminate everything; possibly water she had drank)

6. Young couple with 3-yr old child; travel to endemic country for 2 months → child with 105-106 fevers, high RR, WBC 25
• All diagnostics negative (malaria, sputum, urine, blood), negative CXR
• No response to empiric broad spectrum abx over 3 days → Chest CT = left lobe infiltrate
• **Mycobacteria/Nocardia/fungi/malignancy** on differential when pt still acutely ill with no obvious cause
  o Sputum acid-fast smear microscopy negative; molecular test negative, culture will take 8 wks total to come back
• Start TB therapy → 4 wks later fever persists, culture still negative
  o Could switch to meds for MDR-TB, but could get more tests → gastric aspirates grow TB 2 weeks later and it IS XDR-TB (also resistant to second line TB)
Zoonoses

Zoonoses = any infectious disease that may be transmitted from animals to humans or vice versa
- Account for 60% of infectious diseases in humans (billions of cases, millions of death each year)
- Categorized by route of transmission, pathogen type, degree of person-to-person transmissibility

**For zoonoses, know transmission and clinical manifestations for each disease

Vector = any animals that transmits an agent of human disease or plays an essential role in the agent’s life cycle (typically refers to athropods like mosquitoes, ticks, etc. and less commonly to animal reservoirs)

Emerging Infection = Diseases that have recently appeared or that are increasing in incidence
- Risk factors for emergence = zoonotic, vector borne, bacterial/fungal infections

Transmission of Zoonoses:
- Contaminated food → Listeria, salmonella, Campylobacter, E. Coli, botulism, seafood-borne disease (e.g. Vibrio), Staph food poisoning
- Direct contact with animals or infected materials → leptospirosis, brucellosis
- Animal bites/scratches → cat scratch disease, rat bite disease
- Arthropod vectors (ticks, lice, mosquitoes, etc.) → dengue, Rocky Mountain spotted fever, Tularemia

Leptospirosis → caused by Leptospira species (a spirochete)

Epidemiology
- >10 million infections/year; world-wide distribution (abundant in tropics/rainy season) → “neglected disease” in urban slums and rural farmers
- An emerging infection (due to travel/globalization, water sports, climatic events like floods)

Pathogenesis → Colonizes renal tubules and is shed in the urine (domestic animals and livestock)
- Can survive in the environment for weeks/months
- Contaminated water/soil enters skin abrasions or conjunctivae

Clinical Features → Incubation 2-30 days; acute fever, HA, myalgias, abdominal pain, conjunctival suffusion (red eyes)
- Some develop second (immune) complications → aseptic meningitis, uveitis
- Severe complications
  - Weil’s Syndrome (5-10% pt’s) → renal failure, pulmonary hemorrhage, cardiac arrhythmia, multi-organ failure, death in 5-15%
  - De novo hemorrhage syndrome (>70% mortality)

Diagnosis → Gold standard = MAT (microagglutination test) detects Leptospira species antibodies
- Culture only used during 1st week (acute spirochetemia)
- Darkfield microscopy isn’t very sensitive; immunohistochemistry relatively unavailable
- PCR more sensitive than culture

Treatment → PCN, Cefotaxime, Doxycyline; supportive care for Jarisch Herxheimer reaction

Brucellosis
- Caused by Brucella species (gram-neg coccobacillus, facultative intracellular bacteria)
- 4 main species = B. melitensis (goat/sheep), B. abortus (cattle), B. suis (pigs), B.canis (dogs)
  - B. melitensis and abortus cause more human disease

Epidemiology → partial reporting, but distributed world-wide (endemic Eurasia, Africa, S America)
- Transmission mostly from ingestion of unpasteurized dairy products, but also from inoculation and inhalation
Clinical Features

1. **Classical febrile brucellosis** → acute infection with fever, sweating, malaise, HA, weight loss, arthralgias/myalgias, back pain (*mimic TB or infectious endocarditis*)

2. **Relapsing or undulant brucellosis (Malta fever)** → occurs >2 months after classical infection
   a. Liver involvement, arthritis, uveitis, orchiepididymitis
   i. Osteoarticular Disease → peripheral arthritis, sacroiliitis, spondylitis
   b. May be chronic, lasting over one year

Complications

- Reproductive system → epididymoorchitis (granulomatous inflammation), abortion
- Liver → hepatomegaly, granulomatous hepatitis (caseating or non-caseating)
- CNS → meningitis, meningoencephalitis, encephalitis, brain abscess → bad prognosis
- Endocarditis → Main cause of mortality, usually aortic valve (generally requires surgery)
- Relapse → After inadequate treatment, usually within one year

Diagnosis

- Blood or bone marrow culture → biosafety level 3 lab
- Detection of antibodies

Treatment → Doxycycline PLUS rifampin OR streptomycin for 4-6 weeks

**Q Fever** → Caused by *Coxiella burnetii* (intracellular, especially in phagocytes), found worldwide

Epidemiology

- Reservoir in **cattle, sheep, and goats** → shed in urine/feces; survives on hay, straw, clothes
- Transmission via **inhalation**, inoculation, ingestion → human-to-human is rare
- Incubation period 2-3 weeks; acute and chronic illness possible (only 50% are symptomatic)
  o Only 1 organism required for infection!

Clinical Features → High fever for 1-2 weeks, fatigue, chills, HA, myalgias, sweats, cough, N/V/D, CP, rash

- Chronic Q fever is rare (<5% of pts) → develop sx’s years after symptomatic or asymptomatic infection → is an important cause of culture-negative endocarditis

Diagnosis → Lab findings are non-specific

- Acute infection → PCR or serological titers (4-fold rise), Phase 2 antibody
- Chronic infection → Phase 1 IgG antibody

Treatment → Doxycycline is the drug of choice; pt’s are monitored, hydroxychloroquine might be added

Prevention → inactivated vaccine available in Australia; education to high-risk

**Vector-Borne Zoonoses**

**Dengue Fever** → most important arbovirus infecting humans (1/3 world’s population at risk)

- Endemic in Puerto Rico and many tourist locations in S America, SE Asia, the Pacific
- Spread by *Aedes* mosquito vector (day-biting) → 4 serotypes (humans are natural host)

Epidemiology → rising numbers each year, and more countries reporting it (global pandemic)

- Carrier mosquitoes are moving, also influenced by climate change
- More severe symptoms/form than was seen 50 years ago

Clinical Features → frequently asymptomatic in young children; in older children and adults presents as a nonspecific febrile illness (fever, arthralgia, rash)

- Classically has hemorrhagic symptoms ranging from minor bleeding (purpura, petechiae) to “plasma leak syndrome”, “dengue shock” and vascular collapse
- **Severe dengue** (formerly hemorrhagic dengue) = major bleeding, shock, organ failure
- Complications → vascular (can lead to shock), thrombocytopenia, bleeding

Diagnosis → PCR or cell culture in the beginning of infection (viremia only for several days); then ELISA to assay for IgG and then IgM

Tick-Borne Rickettsial Diseases (TBRD) → all vector-borne; bacteria are all intracellular
- Genera Rickettsia, Ehrlichia, and Anaplasma → obligate intracellular bacteria in anthropod host
  - Rickettsia = cytoplasmic
    - Infect endothelial cells; prevent lysosome fusion in macrophages
  - Ehrlichia and Anaplasma = vacuolar
- Contain DNA, RNA, and ribosomes; divide by binary fission; gram-negative cell wall
- Overall diagnosis is difficult due to intracellular bacteria, often not suspected or reported

**Rocky Mountain Spotted Fever** → Caused by Rickettsia rickettsia

**Epidemiology** → Spread via multiple tick vectors (most common in SE US = American dog tick)

**Clinical Symptoms** → Fever, HA, myalgias, rash (macular, papular, petechial)
- GI, renal (acute tubular necrosis 2° hypotension), cardiopulm (pulm edema), CNS sx's (meningoencephalitis; cerebral edema and herniation)

Diagnosis → Skin biopsy showing R. rickettsi antigen, or PCR on skin biopsy for active infection
- Serological confirmation isn't useful days 7-14 (IgG seroconversion or 4-fold rise in titer)
- Risk of death increases 5x if diagnosed after day 5 of illness → usually not treated until after day 5 due to ineffective diagnosis
  - Typical rash might be absent or develop later, less suspected in off-peak tick season

**Treatment** → doxycycline and tetracycline in adults and kids

**Tularemia** → caused by Francisella tularensis

**Epidemiology** → Disease of wild mammals; transmitted to humans via direct contact with skin abrasion (game like rabbits), deer fly/tick bites → NO person-person spread
- Seen mostly in summer, across most of US, in mostly the very young and older adults

**Pathogenesis**
- 2 types → Type A is more virulent than B
- Virulence linked to ability to multiply within several cell types → growth is unimpeded until phagocytes are encountered, then resists lysosomal fusion → forms ulcerative lesion
  - Natural infection confers long-lasting immunity

**Clinical Symptoms** → Acute fever, chills, malaise; may progress to systemic multi-organ infection
- CDC case definition = ulceroglandular (skin lesion), glandular, ocuglandular, oropharyngeal, intestinal, pneumonial (low inoculum; inhaled), typhoidal (high inoculum)

Diagnosis → Elevated serum antibody titer (+ 4-fold increase) to F. tularensis antigen in a patient w/o history of vaccine OR isolation of F. tularensis from a clinical specimen
- Gram-neg coccobacilli that grow on Chocolate and cysteine-glucose blood agar

**Treatment** → Gentamicin or streptomycin + doxycycline (cipro/chloramphenicol are alternatives)
- Mortality without treatment = 60% for pneumonic, 2% for ulceroglandular
- Live vaccine available for those at unavoidable risk (vets, etc.)
Summary Chart for Non-Vector Borne Zoonoses

<table>
<thead>
<tr>
<th></th>
<th>Leptospirosis</th>
<th>Brucellosis</th>
<th>Q Fever</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Causative Agent</strong></td>
<td>Spirochete - <em>Leptospira</em></td>
<td><em>Brucella</em> species</td>
<td><em>Coxiella burnetii</em></td>
</tr>
<tr>
<td><strong>Transmission</strong></td>
<td>Water-borne via infected urine</td>
<td>Ingestion</td>
<td>Inhalation</td>
</tr>
<tr>
<td><strong>Clinical Symptoms</strong></td>
<td>Febrile illness, sepsis ± jaundice, pulmonary hemorrhage, renal failure</td>
<td>Fever, arthritis, orchitis, abortion, endocarditis, meningitis</td>
<td>Acute Q = nonspecific febrile illness (Phase 1 antibodies) Chronic Q = culture-negative endocarditis (Phase 2 Ab's)</td>
</tr>
<tr>
<td><strong>Treatment</strong></td>
<td>PCN, ceftriaxone, doxy</td>
<td>4-6 weeks of abx; Doxy + rifampin OR streptomycin</td>
<td>Doxycycline</td>
</tr>
<tr>
<td><strong>Other</strong></td>
<td>Histopath = granulomatous inflammation</td>
<td>Intracellular bacteria, esp in phagocytes</td>
<td></td>
</tr>
</tbody>
</table>

Summary for Vector-Borne Zoonoses

<table>
<thead>
<tr>
<th></th>
<th>Dengue Fever</th>
<th>Rocky Mounting Spotted Fever</th>
<th>Tularemia</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Causative Agent</strong></td>
<td>Arboviruses</td>
<td><em>Rickettsia rickettsii</em></td>
<td><em>Francisella tularensis</em></td>
</tr>
<tr>
<td><strong>Transmission</strong></td>
<td>Aedes mosquito bites</td>
<td>Tick bites</td>
<td>Direct contact with abrasion (rabbits/game) or tick bite</td>
</tr>
<tr>
<td><strong>Clinical Symptoms</strong></td>
<td>Asymptomatic, DF, DHF- DSS, leukemia w/o thrombocytopenia → 2nd, 3rd infections are worse than 1st</td>
<td>High fever, HA, rash after 3-5 days, nml WBC with left shift; thrombocytopenia; hypotension and end organ ischemic injury</td>
<td>Mostly ulceroglandular form (local necrotic ulcer)</td>
</tr>
<tr>
<td><strong>Treatment</strong></td>
<td>Supportive</td>
<td>Doxycycline, tetracycline</td>
<td>Aminoglycoside with doxycycline</td>
</tr>
<tr>
<td><strong>Other</strong></td>
<td>Emerging pandemic</td>
<td>Case fatality 8-10%</td>
<td>Type A more virulent than B</td>
</tr>
</tbody>
</table>

Antiviral E-Lecture

Challenges to Antiviral Drug Development → variability in viral replication, drug safety, and emergency drug-resistant viral strains

Influenza

Virus Characteristics → ssRNA "negative sense" virus; HA needed for viral entry; M2 ion channel needed for viral uncoating, NA cleaves virion, required for release/spread

Amantadine → Block M2, alter HA conformation

MOA:
- Primary effect (occurs early) → blocks M2 channel → only inhibits Influenza A; ion flow through channel is required to reduce pH in virion to uncoat RNA
- Secondary effect (occurs late) → causes premature conformation change in HA protein

Pharmacokinetics → good bioavailability, given PO
Limitations → toxicity (CNS) and high levels of resistance (mutations in M2), no efficacy against Flu B/C

Zanamavir (Relenza) & Oseltamivir (Tamiflu) → NA inhibitors
MOA → Mimics sialic acid binds the active site of NA more tightly; preventing sialic acid cleavage/virus release; works against Influenza A and B
Pharmacokinetics → good bioavailability, given PO, excreted by kidneys
Efficacy/Usage → used for both treatment and prophylaxis
  • Reduces durations of illness by 1-2 days only if started within 48 hours of symptom onset
  • Reduced incidence in close contact when used as prophylaxis (70-90%)
Limitations = Resistance (NA mutations) & toxicity → N/V (Oseltamavir), Bronchospasm (Zanamivir)

Herpesviruses
Herpesvirus Characteristics → large, linear dsDNA; 8 different types infect people
  • All of the Herpesvirus drugs act at the level of replication, target the viral DNA polymerase

Acyclovir
MOA → Analog of nucleoside deoxyguanosine, but lacks OH group (chain terminator, stops replication)
  • In order to work, acyclovir must be phosphorylated by viral tyrosine kinase (CMV doesn't have kinase → works against HSV, but not CMV)
Pharmacokinetics → given oral, IV, or topical; poor PO bioavailability and short half-life (dosing often)
  • Valacyclovir is a prodrug with increase bioavailability and dosed less frequently
  • Cleared renally
Toxicity → Well tolerated usually, can cause N/V → renal toxicity with high doses
Efficacy → Effective against HSV or VZV; not CMV/EBV OR latent HSV (but can suppress outbreaks)
Resistance → a problem, mostly in HIV patients (usually due to viral TK mutation, which confer cross-resistance to analogs, including ganciclovir)

Ganciclovir → improved activity against CMV (which has no viral TK to activate acyclovir)
MOA → Similar to acyclovir; also a deoxyguanosine analog and need to be phosphorylation
  • CMV viral protein kinase has affinity for ganciclovir, allows it to be phosphorylated
Efficacy → CMV, HSV, VZV (first line for CMV primarily in HIV and transplant patients)
Pharmacokinetics → Poor PO bioavailability (given IV); a prodrug is available (valganciclovir)
Toxicity → bone marrow suppression (leukopenic, thrombocytopenic); so ganciclovir is reserved primarily for severe infections
Resistance → Mutations in the phosphorylating tyrosine kinase

Foscarnet = second-line agent used when resistance develops to Ganciclovir or Acyclovir
  • MOA → Analog of pyrophosphate → binds polymerase as competitive inhibitor and blocks DNA elongation (doesn't need activation/phosphorylation)
  • Limitations → Only available IV; serious toxicities (renal impairment)
  • Resistance → DNA polymerase mutation

Hepatitis B
  • dsDNA with RNA intermediate used for replication
  • Treatment goal = viral suppression, HBeAg seroconversion, prevent cirrhosis/hepatocellular CA

Interferon
MOA → A cytokine; stimulates host immune system to clear infection; impacts most of life cycle
Pharmacokinetics → Given IM or SC (poor oral availability); usually given as pegylated-IFN-alpha for 48 weeks
Efficacy → 30-70% improvement in virologic response and liver enzymes
Toxicity → Flu-like symptoms are common; bone marrow suppression
**Lamivudine (3TC)** → RT inhibitor; works at the RNA intermediate in Hep B virus replication

**MOA** → Is an analog of cytosine and inhibits RT by acting as a chain terminator

**Pharmacokinetics** → Good PO bioavailability; renal clearance

**Toxicity** → usually well-tolerated with rare side effects

**Efficacy** → 60-70% virologic improvement; poor long-term durability (rebound within a few years when drug is stopped)

**Resistance** → High rates; occurs via mutation of RT (DNA polymerase)

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**Hepatitis C**

**Interferon** → given in combination with Ribavirin; better response for genotype 2 and 3

**Ribavirin** → purine nucleoside analog with broad antiviral activities against RNA viruses

- **MOA** is unclear
- **Given PO** (for HCV) or inhaled (RSV)
- **Toxicity** = anemia, is a significant teratogen

**Telaprevir & Boceprevir**

- **MOA** → Protease inhibitor developed specifically for HCV genotype 1 (most common in US)
  - **Protease** is requires to cleave polyprotein to structural and non-structural proteins
- **Given PO**; well-tolerated; good efficacy (70% with sustained viral response)
- **Resistance due to mutations in the protease** (confers high levels of resistance)

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**SUMMARY OF ANTIVIRAL DRUGS**