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Respiratory Viruses

Respiratory viruses are among the most common causes of disease throughout life. Often mild and self-limited, they are still associated with tremendous economic and societal costs due to school absence, missed work, and physician visits. In addition, inappropriate prescriptions for antibiotics to treat patients with viral upper respiratory infections are a driving force for the growth of antibiotic resistance worldwide. In the developing world, acute respiratory infection (often bacterial pneumonia triggered by viral infections such as influenza) is a leading cause of death in children under age 5. This lecture will cover two of the most important causes of viral respiratory infection, respiratory syncytial virus (RSV) and influenza. It is important to remember that a large number of other viruses, including parainfluenza, human metapneumovirus, and coronaviruses, can cause respiratory infections as well.

Respiratory Syncytial Virus

Epidemiology

RSV infection occurs in essentially all children by the second year of life. Yearly, worldwide epidemics with peaks in the winter months disproportionately affect young children. In the United States, 1-2% of healthy infants are hospitalized for RSV (>50K hospitalizations/year!) While the disease is usually self-limited, there are risk factors for a severe clinical course including prematurity, young age during the RSV season, immunocompromised states, and congenital heart or lung diseases.

Molecular Biology/Pathogenesis/Evolution

RSV is a member of the Paramyxoviridae, an important group of viruses including measles and mumps viruses. These enveloped viruses have a single-stranded, negative-sense RNA genome that encodes 11 proteins. Because it is a negative-strand RNA, viral proteins cannot be made directly from the genome. Thus, the virus must carry an RNA-dependent RNA polymerase that makes positive-sense RNAs for production of viral proteins. This polymerase is associated with the nucleocapsid, a protein coat that is, in turn, surrounded by an envelope derived from the host cell membrane. Two of the most important RSV components are the F and G glycoproteins. These are exposed on the surface of viral particles and mediate attachment to host cells, fusion of infected cells with one another (F protein-dependent, leads to the characteristic "syncytia" seen on microscopy).

RSV replicates well in the cells of the upper airway and is transmitted from person to person by respiratory secretions. It survives well on solid surfaces (including hands and stethoscopes!) and is a leading cause of hospital-acquired infections. Clinically significant RSV disease is caused by spread of the virus to the lower airways -- either the bronchioles, leading to "RSV bronchiolitis" or to the alveoli, leading to pneumonia. Infection of the lower airways is associated with significant epithelial cell necrosis, shedding of dead cells, subsequent air trapping, and atelectasis (collapse of portions of the lung). There is no viremic phase, and pathology is generally confined to the airways.

Clinical Manifestations

Following an asymptomatic incubation period of 3-6 days, affected children initially develop signs of upper respiratory infection. These include rhinorrhea and congestion with or without fever. Some children develop a croup-like illness (laryngotracheobronchitis) with associated cough and stridor. In roughly half of cases, RSV bronchiolitis develops, with infection of the small airways and the clinical signs of cough, prominent wheeze. This is the most common reason for hospital admission due to RSV disease. The duration of illness ranges from 1-3 weeks, and recovery is generally complete. There is, however, an unclear relationship between RSV bronchiolitis in childhood and later development of reactive airway disease (asthma).

A small number of infants may develop frank RSV pneumonia, requiring more intensive monitoring and, sometimes, ventilatory support. Young infants with RSV infections may have episodes of apnea, with an increased risk associated with decreased age.

Older children and adults with pre-existing asthma may develop wheezing in response to reinfection with RSV, and elderly patients may have exacerbations of heart failure during RSV infection. The risk of severe disease is greatly increased among premature illness and those with immune compromise or congenital heart disease.

Prolonged shedding of RSV in children with immunodeficiencies suggests a major role of cell-mediated immunity in clearance of infection. Immunity following infection is incomplete, and reinfections can occur throughout life.

Diagnosis

A compatible clinical syndrome during the winter months is generally sufficient for a diagnosis of RSV bronchiolitis. In hospitalized cases, a specific viral diagnosis, either by viral culture or by rapid antigen detection, is often sought. This is useful both for predicting clinical course and for facilitating hospital infection control measures.

Treatment

There is no specific antiviral treatment for RSV, and there is no vaccine. Treatment of RSV disease is generally supportive, with young infants more likely to require hospitalization and monitoring. Severe cases may require intubation and mechanical ventilation. Steroids and bronchodilators are of unclear utility. Ribavirin, which has some activity against RSV, is used in severe cases with prolonged viral shedding. It's utility is, to be kind, unclear.

Prevention

The cornerstone of RSV prevention is through handwashing and cleaning of potential fomites in situations (such as day care) in which RSV is likely to spread. Hospitalized cases are placed on "contact isolation" (gowns/gloves) to prevent spread within the facility.

Antibody-based prophylaxis is useful in high risk infants (prematurity, chronic lung disease, congenital heart disease). This consists of monthly doses of palivizumab (a

humanized monoclonal antibody against the F glycoprotein) during RSV season. Palivizumab represents an improvement over the prior prophylaxis strategy, an RSV hyperimmune globulin preparation (RSV-IGIV). Vaccine strategies have been disappointing to date.

Influenza

Epidemiology

The recent emergence of a novel, swine-associated influenza A strain that has resulted in a pandemic makes this an interesting time to discuss the influenza virus. Despite the availability of an effective vaccine, annual seasonal outbreaks of influenza lead to a tremendous burden of disease and significant morbidity and mortality worldwide. In the U.S., there are more than 200,000 hospitalizations and 36,000 deaths due to influenza every year.

Clinical Manifestations

Influenza ("the flu") is usually a self-limited illness associated with fever, chills, headache, myalgias, arthralgias, and cough. Young children may have an atypical presentation with a predominance of gastrointestinal symptoms (this has been noted during the current H1N1 pandemic). The potential complications of influenza infection are quite significant and include bacterial pneumonia and/or sepsis. Myositis (muscle inflammation) may occur during influenza infection, especially with type B viruses. Other rare complications include encephalitis and (in children receiving aspirin therapy) Reye syndrome (liver damage and encephalopathy).

The incubation period of influenza is 3-5 days, and the duration of uncomplicated influenza is generally less than a week.

Molecular Biology/Pathogenesis/Evolution

Influenza viruses are members of the *Orthomyxoviridae*. These are enveloped viruses with a segmented, negative-sense RNA genome. The genomes of influenza A and B viruses have 8 segments, and influenza C (not a significant cause of disease) has 7, as it lacks a neuraminidase gene. Viral proteins encoded by these segments include:

HA = hemagglutinin
NA = neuraminidase (influenza A, B only)
PB1, PB2, PA = polymerases
NP = nucleocapsid
L = large polymerase protein
M1 = associated with NP
M2 = ion channel (influenza A only)
NS = non-structural proteins

The hemagglutinin and the neuraminidase are the immunodominant antigens and are the major determinants of the viral serotype (e.g. H1N1 vs. H3N2). The hemagglutinin is important for epithelial cell attachment and entry, and the neuraminidase is important for viral budding from infected cells. Following attachment, the virus enters the cell via endocytosis. The low pH of the endosome leads to a change in the conformation of viral

envelope glycoproteins and escape of viral material into the cytoplasm. Genomic RNA translocates to the nucleus, where the viral RNA-dependent RNA polymerases transcribe positive-sense RNAs used for making viral proteins and host RNA polymerase is co-opted for transcription of negative-sense viral genomes that are packaged into new virions. The dsRNA intermediates formed during RNA-dependent RNA polymerase activity are called "replicative intermediates."

Influenza viruses infect the columnar epithelial cells of the respiratory tract, leading to epithelial destruction, increased mucus production, and ciliary stasis. Replication is maximum within about 3 days of infection, but virus may be shed for up to 7-10 days in uncomplicated influenza (much longer in immunocompromised patients). Both innate and adaptive immune responses appear to be important for clearance.

Nomenclature: Type/host species (human default)/location/year (HA/NA type)
Influenza A/California/2009 (H1N1)

Antigenic shift and drift are important concepts in influenza biology and are very relevant to the spread of novel strains. **Antigenic drift** is due to the accumulation of mutations in the RNA genome encoding the HA and NA proteins. RNA genomes lack many of the proofreading functions of DNA genomes, so the development of mutations is rapid. Most mutations are deleterious (i.e. will lead to less fit viruses and will be selected out of populations). However, some changes may lead to enhanced replication or better evasion of host immune responses. These may lead to changes in the population of circulating viruses over time and are, in large part, the reason for the limited cross-protection against influenza from season to season. **Antigenic shift** is the result of the segmented nature of the influenza genome. Cells may be infected with multiple types of influenza virus simultaneously. Some animals (pigs, poultry) may be particularly permissive to coinfection by influenza strains. Because the genomes are segmented, reassortment of segments may occur during viral packaging and release. Again, most of these will lead to less fit viruses. At times, though, the result can be a virus that can infect and be contagious in humans and to which the population has little pre-existing immunity. These viruses are the ones with the potential to sweep rapidly through human populations. This is thought to have been the mechanism for the emergence of the 1918 H1N1 influenza that killed more than 20 million people. The currently circulating pandemic "swine influenza" appears to be related to a triple-reassortant virus with genome segments from swine, avian, and human strains.

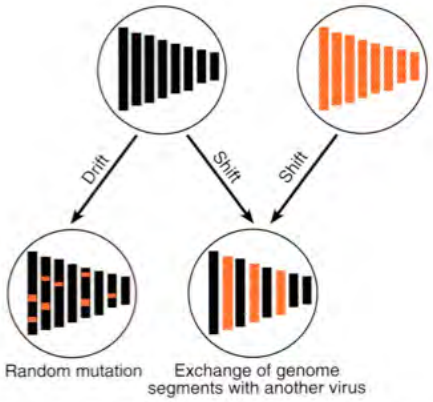

Figure 1.20: Principles of antigenic drift and shift. Colored bars represent novel RNA sequences.

Diagnosis

The diagnosis of influenza is suggested by a compatible clinical syndrome during a seasonal epidemic, but it is important to remember that many other viruses can cause "flu-like" illnesses. Viral culture or rapid antigen testing from nasal secretions can confirm the diagnosis of influenza. PCR testing has been widely deployed during the novel H1N1 outbreak but is still not generally used for seasonal influenza.

Treatment

Treatment of uncomplicated influenza is supportive; however, specific antiviral agents exist for patients at high risk of complications. The adamantanes (amantidine, rimantidine) are primary symmetric amines that block the activity of the influenza A M2 protein (an ion channel that promotes viral uncoating). They have no activity against influenza B. If given in the first 48 hours of symptoms, amantidine may shorten the course of the disease by a day or two. In addition, there is a role for both drugs as post-exposure prophylaxis in outbreak settings. The neuraminidase inhibitors (oseltamivir, zanamivir) can also be used as treatment (most effective within 72 hours of symptom onset) or prophylaxis. Resistance is emerging to these agents, including among some isolates of the novel H1N1 influenza.

Prevention

The mainstay of prevention of seasonal influenza is the influenza vaccine. Because of antigenic drift and circulation of new viral types each year, the influenza vaccine formulation changes annually. Influenza vaccine contains two type A strains and one type B strain each year. Viruses are grown in eggs (and people with anaphylactic reactions to egg proteins cannot receive influenza vaccine). The trivalent, inactivated vaccine (TIV) is a formalin-killed mixture of vaccine strains given intramuscularly. A live-attenuated intranasal vaccine (LAIV) consists of recombinant viruses that have 6 genes from a cold-adapted influenza in combination with HA and NA genes from that year's circulating strains. It is used in healthy individuals aged 5-49 and is associated with a small risk of wheezing due to viral replication. Adults and older children get a single annual dose of either vaccine preparation, but children under age 9 require two doses in the first season that they receive vaccine. Vaccine efficacy is estimated at 50-80%, and it significantly decreases the risk of severe disease and hospitalization in targeted populations.

Who gets influenza vaccine?

- You! (health care workers)
- Children 6 months – 18 years; adults > age 50
- Contacts of children < 6 months or other high risk people
- Pregnant women
- Chronic medical conditions
- Aspirin therapy
- Chronic care facilities
- Anyone who wants it!

Who doesn't get influenza vaccine?

- Inactivated vaccine (TIV)
    - Children under 6 months
    - Anaphylactic reaction to eggs or other vaccine components

- Live-attenuated vaccine (LAIV)
    - Children less than 5 years or adults > 50 years
    - Anaphylactic reaction to eggs or other vaccine components
    - Immunocompromised or other high risk for severe influenza
    - Currently receiving salicylates (aspirin)
    - History of Guillain-Barré syndrome
    - Asthma
    - Pregnant women