Chapter 39

RNA viruses (1)

RNA enveloped viruses:

Orthomyxoviruses

Influenza viruses

-- Influenza viruses are **important human pathogens**.

-- They cause **outbreaks** of influenza that **sicken and kill thousands of people every year** as well as **infrequent pandemics**.

-- They are the **only member** of this family which differs from paramyxoviruses primarily in that it has a **segmented RNA genome (usually 8 pieces)** whereas **paramyxoviruses have a single piece**.

-- Orthomyxoviruses are **smaller** than paramyxoviruses.

-- **Myxo** refers to interaction with **mucin**.

Human influenza virus

-- **Influenza A** virus causes **pandemics**.

-- **Influenza B virus** causes **major outbreaks** and **influenza C virus** causes **mild respiratory tract infections**.

-- **Pandemics** occur when a **variant of influenza A virus** that contains a **new haemagglutinin** against which **people do not have preexisting antibodies** is introduced into the human population.

-- Pandemics occur **infrequently** but major outbreaks occur almost every year.

-- Outbreaks of **influenza B virus** occur less frequently.

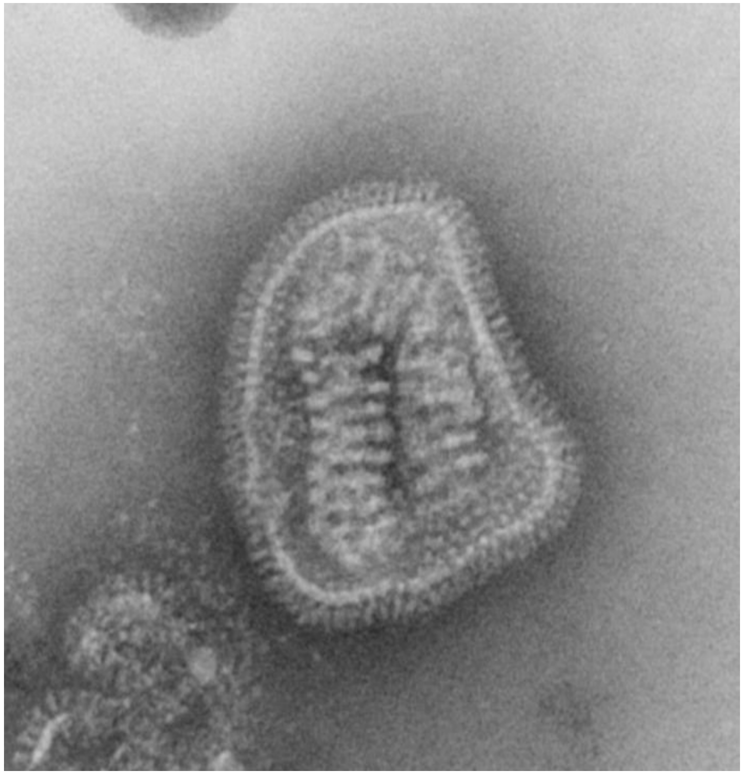
Important properties

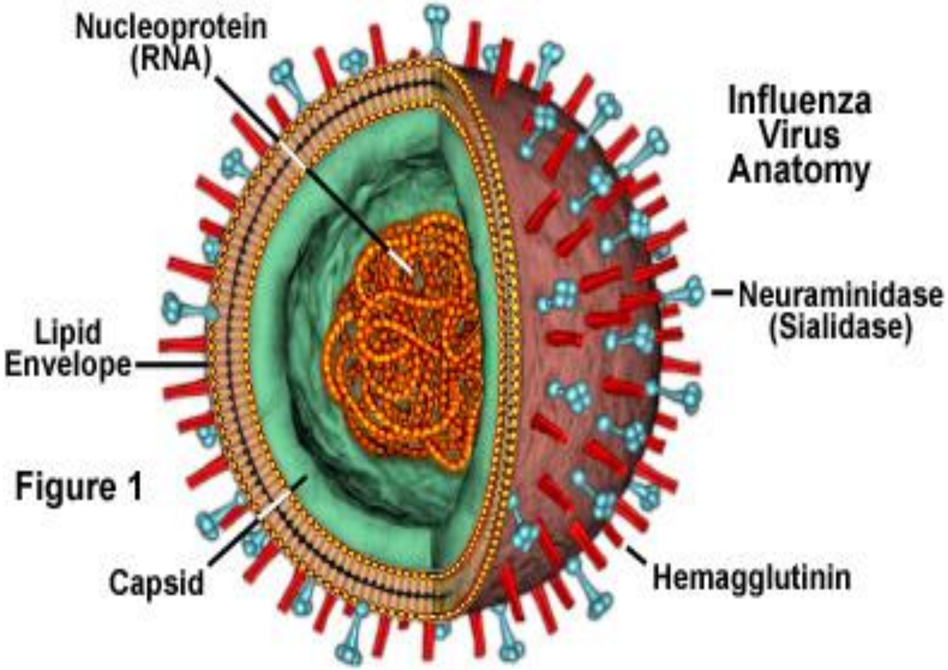
It is composed of a **segmented single stranded RNA genome**, a **helical capsid** and an **outer lipoprotein envelope.**

Virus contains **RNA dependent RNA polymerase** which **transcribes the negative polarity genome into mRNA.**

Envelope has **two different spikes**, a **hemagglutinin (HA)** and a **neuraminidase (NA).**

Influenza A virus has **16** antigenically distinct types of **HA** and **9** of **NA**.




Nucleoprotein
(RNA)
Influenza
Virus
Anatomy
Neuraminidase
(Sialidase)
Lipid
Envelope
Figure 1
Capsid
Hemagglutinin

- **HA** bind to cell surface receptors **neuraminic acid (sialic acid)** to initiate infection.

- **NA** cleaves neuraminic acid to **release progeny virus** from infected cell.

- It also **degrades protective layer of mucus** to enhance the ability of virus to gain access to the respiratory epithelial cells.

- **Ability of influenza virus to change antigenicity of HA and NA contributes to devastating pandemics.**

These antigenic changes are of two types:

Antigenic shift

-- It is based on **reassortment of segments of RNA genome**.

-- **Entire segments are exchanged**, each of which **code for a single protein**.

-- It is the major change.

Antigenic drift

-- It is based on **mutations in the RNA genome**.

-- **Influenza A virus** has **two matrix proteins**.

-- **M1** is **located between the internal nucleoprotein and the envelope**, and provides **structural integrity**.

-- **M2** forms an **ion channel between the interior of the virus and the external milieu**.

-- This ion channel plays an essential role in the **uncoating of the virion** after it enters the cell.

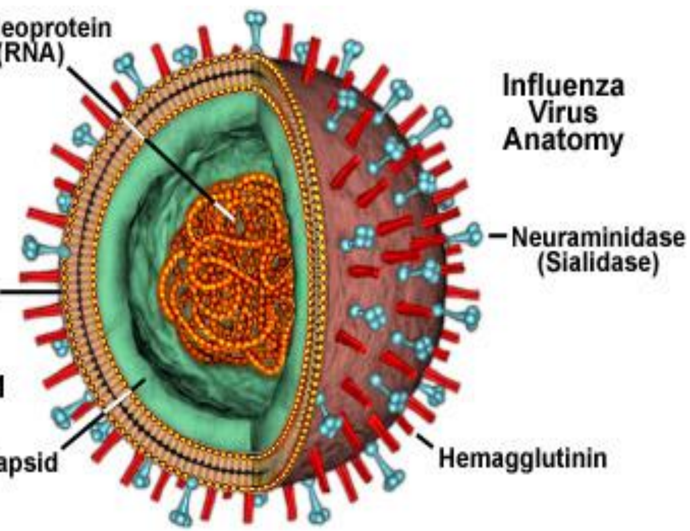


Figure 1

Influenza virus has both **group specific** and **type specific** antigens:

1. The **internal ribonucleoprotein** is the **group specific antigen** that distinguish **A, B,** and **C** viruses.

2. **HA** and **NA** are **type specific surface proteins**. **Antibody against HA neutralizes infectivity** and prevent disease. **Antibody against NA** does not neutralize infectivity but **reduce disease severity** by decreasing amount of viruses released from infected cell.

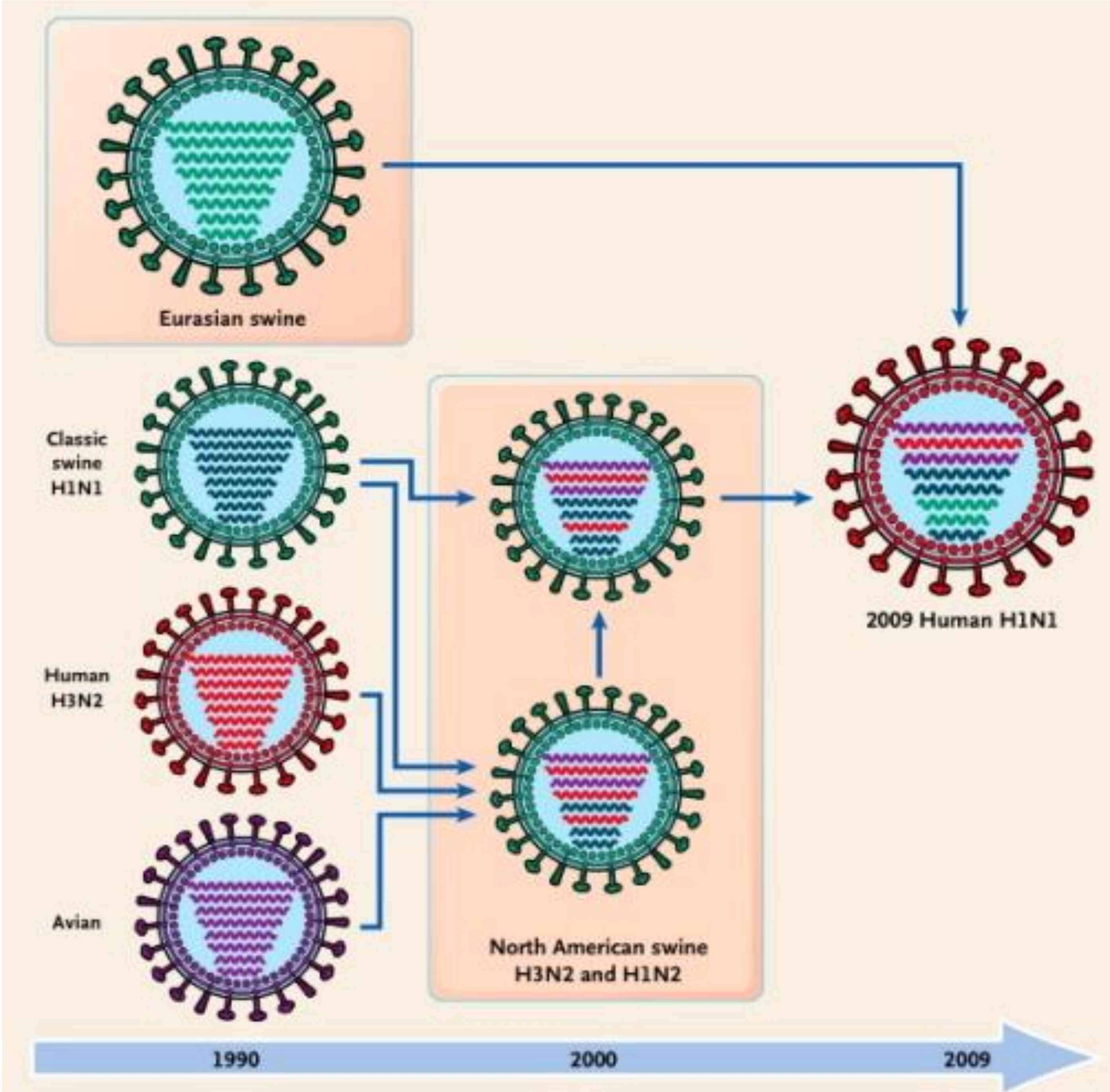
3. A **nonstructural protein NS1** is an important determinant of **virulence**. It **inhibits interferon** production.

-- Many **animal species** have their **own influenza A viruses** which are source of antigenic shift in human virus.

For example, if an **avian** and a **human** influenza A virus **infect the same cell**, **reassortmen**t could occur and a **new variant** of the human A virus, bearing the avian virus HA may appear.

-- **Birds** are a common source of such genes.

-- **Pigs** may serve as the **“mixing bowl”** within which the human, avian, and swine viruses reassort.


Eurasian swine
Classic swine H1N1
Human H3N2
Avian
North American swine H3N2 and H1N2
2009 Human H1N1
1990
2000
2009

-- **Influenza B virus** is a **strict human virus** so it **does not undergo antigenic shifts**.

-- But there are **sufficient antigenic drifts** to produce a new version every year.

-- It has no antigens in common with influenza A.

Nomenclature can be understood by following example:

-- In **A/Philippines/82 (H3N2)**, **A** represents **influenza A virus**, then the country of origin followed by year. In brackets are the **HA** and **NA** types.

-- Currently **H1N1** and **H3N2** strains are most common.

Replicative cycle

-- The **viral haemagglutinin** is modified by **extracellular enzymes** to facilitate its **attachment to receptor** on cell surface.

-- The virus **enters the cell** and **uncoats in an endosome** with **low pH**.

-- **Protons** pass through **ion channels** on viral surface formed by **M2 protein**.

-- This **disrupts the envelope** and nucleocapsid is released to **enter cell nucleus via cytoplasm** where 8 segments of RNA genome are **transcribed** into 8 distinct mRNAs.

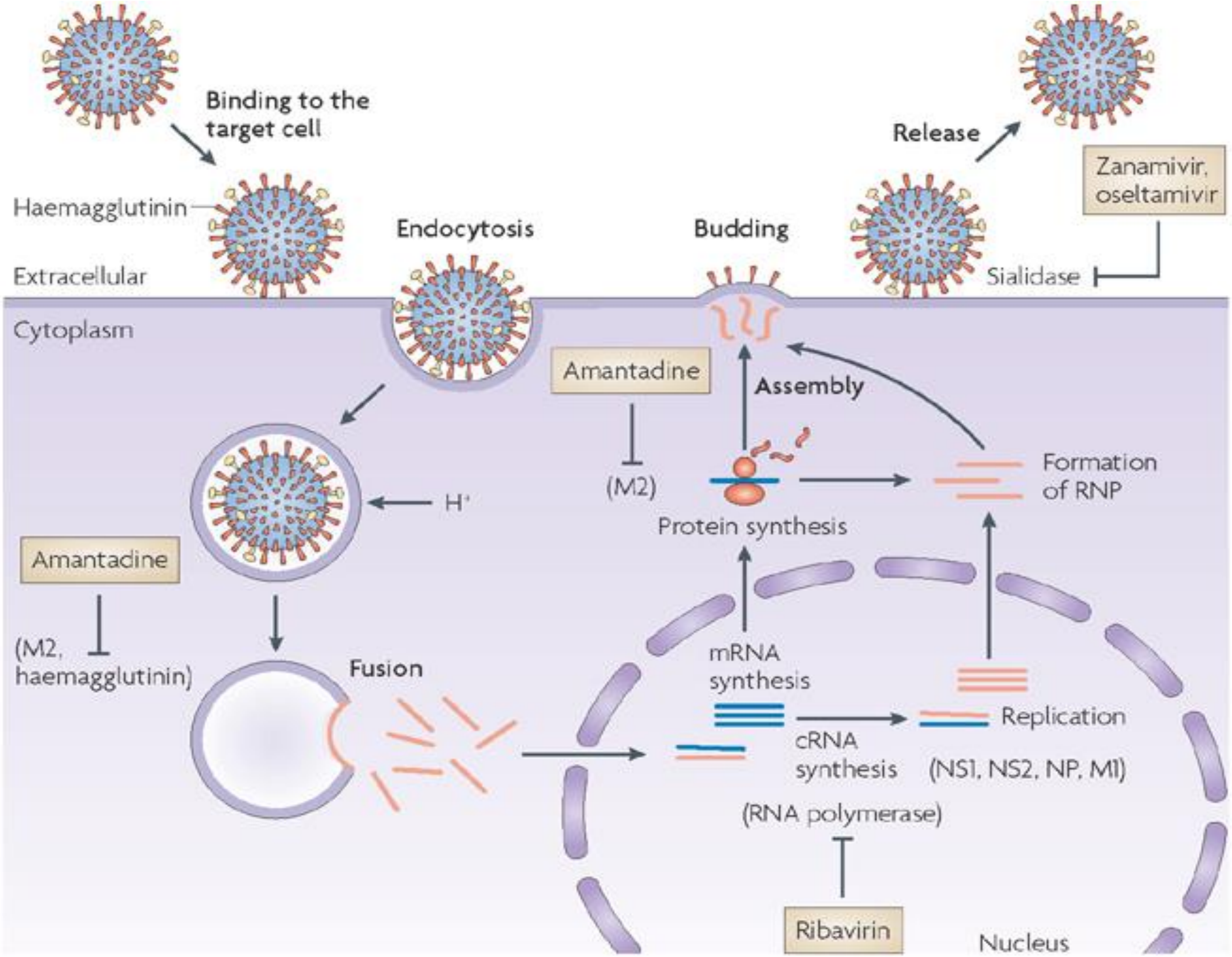
-- **Most** of the mRNAs **move to cytoplasm** where they are **transcribed into proteins**.

-- Some **mRNAs remain in nucleus** to serve **as template for synthesis of negative strand RNA genomes** for progeny viruses.

-- Two newly synthesized proteins **NP protein** and **matrix protein bind to progeny RNA genome in nucleus** and that **complex is transported to the cytoplasm**.

-- The **helical ribonucleoprotein assembles in the cytoplasm** and interact with envelope through matrix proteins.

-- Virions are released from outer cell membrane by **budding**.



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Transmission and epidemiology

-- Transmission is by **airborne respiratory droplets**.

-- **Antigenic shift variants appear infrequently** whereas **drift variants appear virtually every year**.

-- Last pandemic was in **1968** when **H3N2** emerged.

-- These occur when antigenic shift has become so significant that existing immunity in a community can no longer counter it.

-- Influenza occurs primarily in winter.

-- **Along with secondary bacterial pneumonia**, it causes a significant number of deaths especially in **older people**, and **children younger than 2 years**.

Pathogenesis and immunity

-- Infection is limited primarily to **respiratory tract**.

-- **Viremia** rarely occurs.

-- There is necrosis of superficial layers of respiratory epithelium.

-- Influenza virus pneumonia is **interstitial** in location.

-- Immunity depends mainly on secretory IgA in the respiratory tract.

-- **IgG** is produced but is **less protective**.

-- **Cytotoxic T cells** also play a protective role.

Clinical findings

-- After **24-48 hours** incubation period, **fever, myalgias, headache, sore throat**, and **cough** develop suddenly.

-- **Severe myalgias** coupled with **respiratory tract symptoms** are typical of influenza.

-- Vomiting and diarrhea are **rare**.

-- The symptoms usually **resolve spontaneously** in 4-7 days, but **influenza or bacterial pneumonia**, esp. by ***Staph. aureus***, may **complicate** the course.

Laboratory Diagnosis

-- Mostly diagnosis is made on **clinical grounds**.

-- Virus can be detected from secretions by **direct fluorescent antibody**, **PCR** or **cell culture**.

-- **Rapid tests** based on **monoclonal antibodies** are also available.

-- These rapid tests help in initiation of treatment with **neuraminidase inhibitors** within 48 hours of onset of symptoms.

-- A **fourfold increase in titer** (**hemagglutination** or **CFT**) with 10 days gap between two samples is diagnostic.

Treatment

-- **Neuramidase inhibitors** are used both for **treatment** and **prevention**.

-- They **prevent spread of infection** and are useful for both **influenza A** and **B viruses**.

-- They are **constantly updated** (just like antibiotics) to encounter new strains.

Prevention

-- **Vaccine** containing two **influenza A virus** strains, e.g., **H1N1** and **H3N2** and one **influenza B virus** strain is commonly used.

-- The **live vaccine** has **temperature sensitive mutants** of **influenza A** and **B viruses**.

-- These mutants **can replicate in the cooler (33°C) nasal mucosa** where they induce IgA **but not in warmer (37°C) lower respiratory tract.**

-- Beside vaccine, prevention can be helped by use of **neuramidase inhibitors.**

-- They are **not a substitute for vaccines** but can be helpful in elderly people.

Avian influenza virus infection in humans

-- **H5N1** strain of **influenza A virus**, which causes **avian influenza** caused an **aggressive form of human influenza** with **high mortality** in Hong Kong in 1997.

-- Humans have its receptors **only in alveoli**, so they are not frequently infected.

-- Its **greater virulence** as compared with H1N1 and H3N2 is due to **relative resistance to interferon** and **increased induction of cytokines** especially **TNF**.

-- There is **no human vaccine**.

Swine influenza virus infection in humans

-- In April 2009, a novel **swine origin strain (S-OIV)** of **influenza A virus** **(H1N1)** originated in Mexico and caused an outbreak of human influenza in **208 countries** by December 2009.

-- There were nearly **10,000** deaths.

-- However, it declined sharply afterwards.

-- It primarily affected **young children**.

-- It **can spread** from human to human.

-- A **PCR** test is now available.

Paramyxoviruses

- **Paramyxoviruses** family contains **four** important members, **measles virus, mumps virus**, **respiratory syncytial virus (RSV)** and **parainfluenza virus**.

-- They **differ from** **orthomyxoviruses** in that their **genomes are not segmented**, **have a larger diameter** and their **surface spikes are different**.

-- They are composed of **one piece of single-stranded RNA**, a **helical nucleocapsid** and **an outer lipoprotein envelope.**

-- The virion contains an **RNA-dependent RNA polymerase** which transcribes the **negative polarity genome into mRNA.**

-- The **envelope is covered with spikes** which contain **hemagglutinin**, **neuraminidase** or a **fusion protein** that causes cell fusion and in some cases, hemolysis.

Measles Virus

It causes measles, a **childhood disease** characterized by **maculopapular rash**.

Important Properties

-- The genome is typical of a **paramyxovirus**.

-- The virion has **two types of envelope spikes**, one with **haemagglutinating activity** and the other with **cell fusing and hemolytic activities**.

-- It has a **single serotype**, and the **haemagglutinin** is the antigen against which neutralizing antibody is directed.

Replicative cycle

-- After **penetration into cell** by haemagglutinin, the virus penetrates and uncoats and virion RNA polymerase transcribes the **negative-strand genome into multiple mRNAs.**

-- Each of these mRNAs is translated into a **specific protein**.

-- Rest of the cycle is **similar** to influenza virus.

Transmission and Epidemiology

-- Transmission is via **respiratory droplets** produced by **coughing and sneezing** both during the prodromal period and a few days after the rash appears.

-- Measles occurs **worldwide** usually in **outbreaks every 2-3 years**.

-- According to WHO, there are **30 million cases each year**.

-- It has one of the **highest attack rates** and can have devastating effect on populations not previously exposed to it.

-- **Malnourishment** especially **Vitamin A deficiency** is a factor.

-- Persons with **deficient CMI** may have serious **life threatening** disease.

Pathogenesis and Immunity

-- Virus enters **blood** after infecting cells of **upper respiratory tract**.

-- From blood it attacks **reticuloendothelial cells** where it replicates again.

-- **Rashes on skin** are caused by **destruction of virus infected cells by killer T lymphocytes**.

-- Soon after rash appearance, patient becomes **non infective** and virus cannot be recovered.

-- **Multinucleated cells** which form due to **fusion protein** in spikes, are characteristic of lesions.

-- **Lifelong immunity** occurs.

-- Although **IgG can play a role in** viral neutralization, **CMI is more important**.

-- **Maternal antibody** protects infant for 6 months.

-- Measles infection may **transiently lower CMI**, allowing infections by other intracellular pathogens such as ***M. tuberculosis*** to **reactivate and flourish**.

-- This effect is due to **binding of virus to its receptor CD46** on surface of **human macrophages** to **suppress IL12 production**.

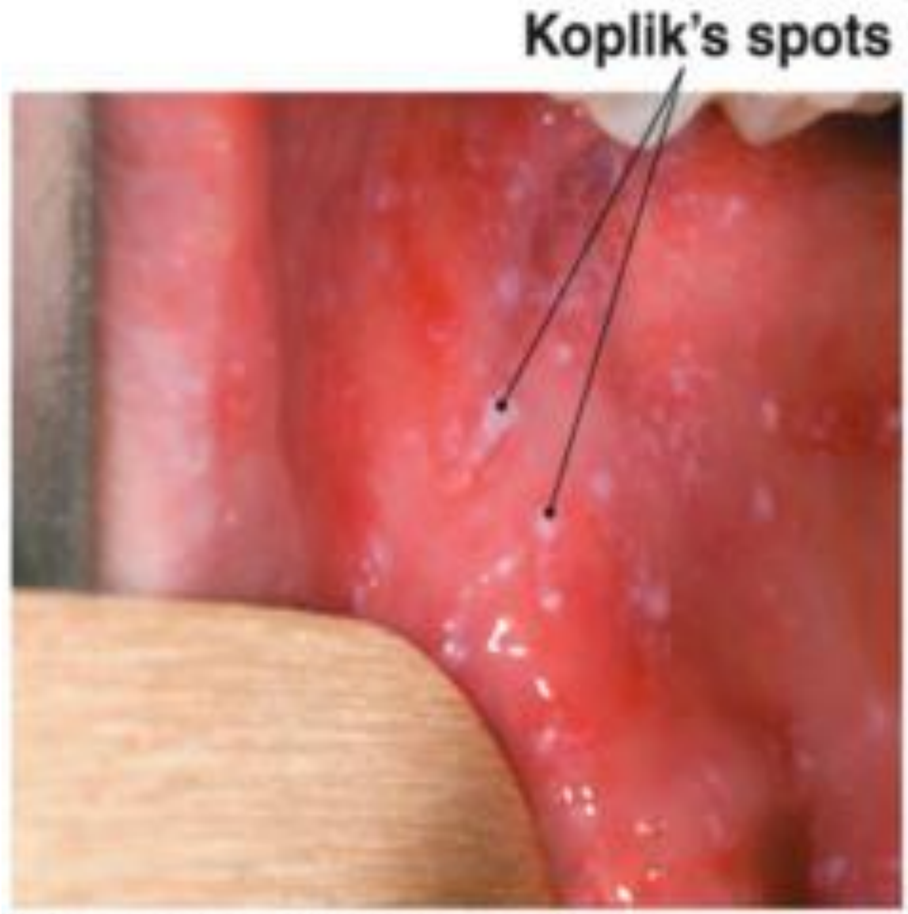
Clinical findings

-- After an incubation period of 10-14 days, a prodromal phase characterized by **fever, conjunctivitis** (causing photophobia), **running nose** and **coughing** occurs.

-- **Koplick's spots** are **bright red lesions with a white central dot** that are located on the **buccal mucosa** and are **virtually diagnostic**.

-- A few days later, a **maculopapular rash** appears on the face and proceeds gradually down the body to **lower extremities**, including palm and soles.

-- The rash develops a **brownish hue** several days later.


Koplik's spots

-- **Complications** of measles can be **very severe**.

-- **Encephalitis** occurs in 1 in 1000 cases with a **mortality of 10%** with permanent sequelae such as **deafness** and **mental retardation** in 40% cases.

-- **Primary measles (giant cell) pneumonia** and **secondary bacterial pneumonia** occur.

-- **Bacterial otitis media** is quite common.

-- ***Subacute sclerosing panencephalitis (SSPE)*** is a rare**, fatal disease of CNS** that occurs several years after measles.

-- Measles in a **pregnant woman** leads to an increased risk of **still birth** rather than congenital abnormalities.

-- Infection of fetus causes death.

-- **Atypical measles** may occur with persons **previously given a killed vaccine**.

-- There is an **atypical rash without Koplik’s spots**.

Laboratory diagnosis

-- Diagnosis is mostly made on **clinical grounds**, but virus can be **isolated in cell culture**.

-- Difficult cases can be diagnosed by **four-fold rise in antibody titer**.

-- **PCR** may also be used.

Treatment

-- There is **no antiviral therapy** available.

Prevention

-- It is based on immunization with **live, attenuated vaccine**.

-- The vaccine is **effective and causes few side effects**.

-- It is given **subcutaneously at 15 months of age** in combination with **rubella** and **mumps** vaccines.

-- It should **not be given before this age** because maternal antibodies may neutralize the virus.

-- **Booster doses** are helpful.

-- Vaccine **should not be given** to immunocompromised persons or pregnant women.

Mumps virus

It is a disease, primarily of **childhood** characterized by **parotid gland swelling**.

Important properties

-- Viral structure is **typical of a paramyxovirus**.

-- There are **two types of spikes**, one with **haemagglutinin and neuraminidase activities** and the other with **cell fusing and hemolytic activities**.

-- There **is single serotype**.

Neutralizing antibody is directed against **haemagglutinin**.

Internal nucleocapsid protein is the **S (soluble) antigen** detected in the CFT.

Surface antigen is called **V (viral)** antigen.

Humans are the **natural hosts**.

Replication is similar to measles virus.

Transmission and Epidemiology

-- Transmission is via **respiratory droplets**.

-- It has **worldwide** occurrence with peak in **winter**.

-- About 30% children have a **subclinical infection** which confers immunity.

Pathogenesis and immunity

-- The virus infects the **upper respiratory tract** and then **spreads through the blood** to infect the **parotid glands, testes, ovaries, pancreas,** and in some cases, **meninges**.

-- **Alternatively**, the virus may **ascend from buccal mucosa up the Stensen's duct** to the **parotid gland**.

-- Disease results in **lifelong immunity**.

-- Unilateral mumps is not followed by mumps on other side.

-- Mumps occurs **only once** and subsequent cases of parotitis can be caused by **parainfluenza viruses**, by **bacteria**, or by **duct stones**.

-- Maternal antibody provides protection for **6 months**.

Clinical findings

After an incubation period of **18-21 days**, a prodromal stage of **fever, malaise,** and **anorexia** is followed by **tender swelling of the parotid glands**, either **unilateral or bilateral**.

There is characteristic increase in **parotid pain** when drinking citrus juices.

The disease is **typically benign** and **resolves spontaneously within one week**.

-- **Complications** include **orchitis** in postpubertal males, which if bilateral can result in **sterility**.

-- **Unilateral orchitis** does not cause sterility.

-- Other complication is **meningitis** which is usually **benign, self-limited** and without sequelae.

Laboratory diagnosis

-- **Diagnosis can be made clinically** but laboratory tests can confirm it.

-- The virus can be **isolated in cell culture** from **saliva, spinal fluid,** or **urine.**

-- **PCR** can also be used.

-- **Four fold rise** in antibody titer (haemagglutination inhibition or CF test) is **diagnostic**.

-- **CFT** may also be used for **S** and **V antigens**.

-- **S positivity** means **current infection**.

-- If **only V is positive**, it indicatives **past infection**.

-- A skin test for **delayed hypersensitivity** may be used to determine **competence of CMI**.

Treatment

-- There is **no antiviral therapy**.

Prevention

-- It is based on a **live, attenuated vaccine**.

-- It is **effective and long lasting** (at least 10 years) with **few side effects**.

-- **Initial does is at 15 months** with a **booster after 4-6 years** usually in combination with rubella and measles vaccine.

-- It **should not be given** to immunocompromised persons or pregnant women.

-- **Ig is not useful in preventing orchitis.**


Thank You!