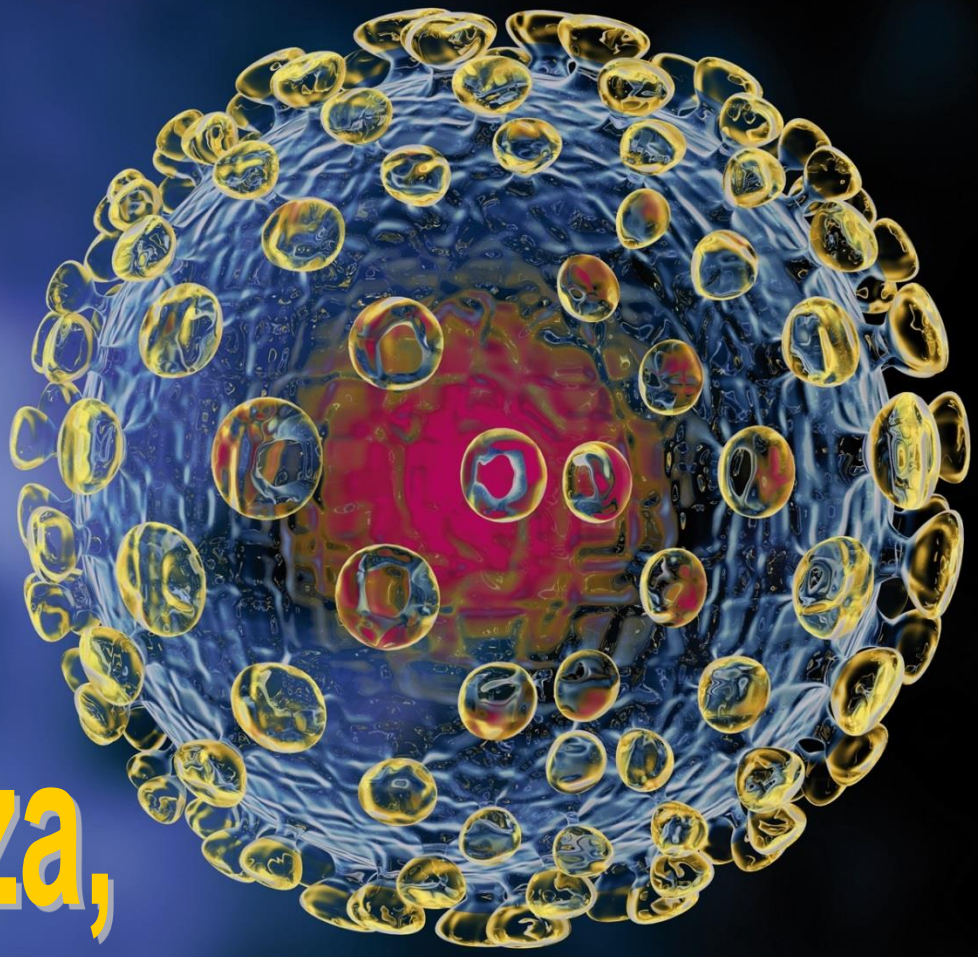
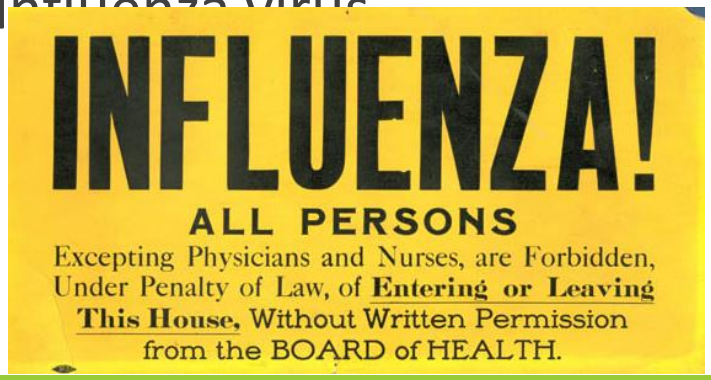
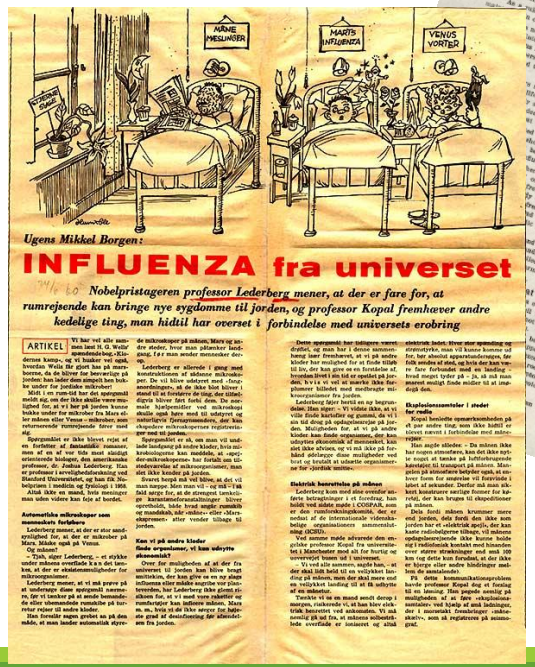


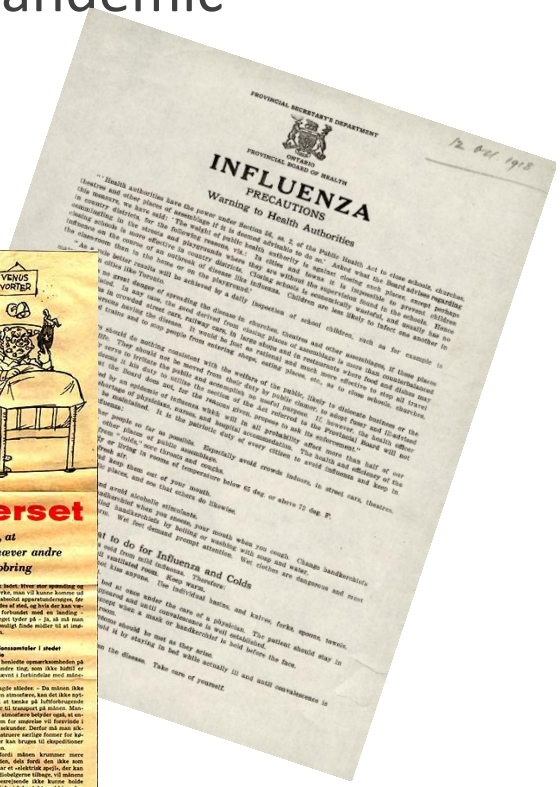
Influenza, Measles and Rubella

Influenza

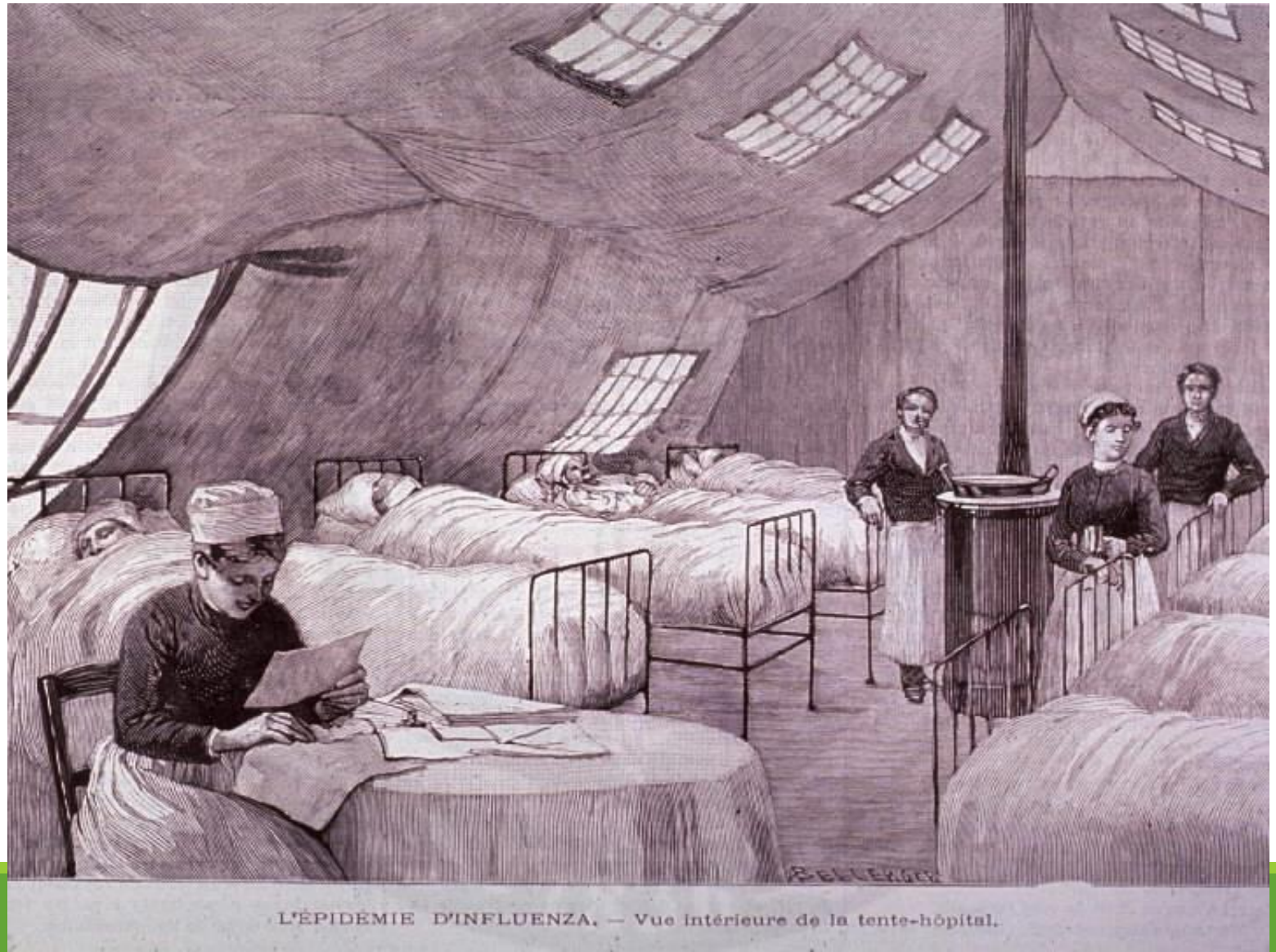
Influenza is the most important of the great epidemic diseases. From time to time, influenza becomes pandemic and sweeps throughout the world. World-wide pandemics of influenza are due to the emergence of antigenically new strains of influenza virus.





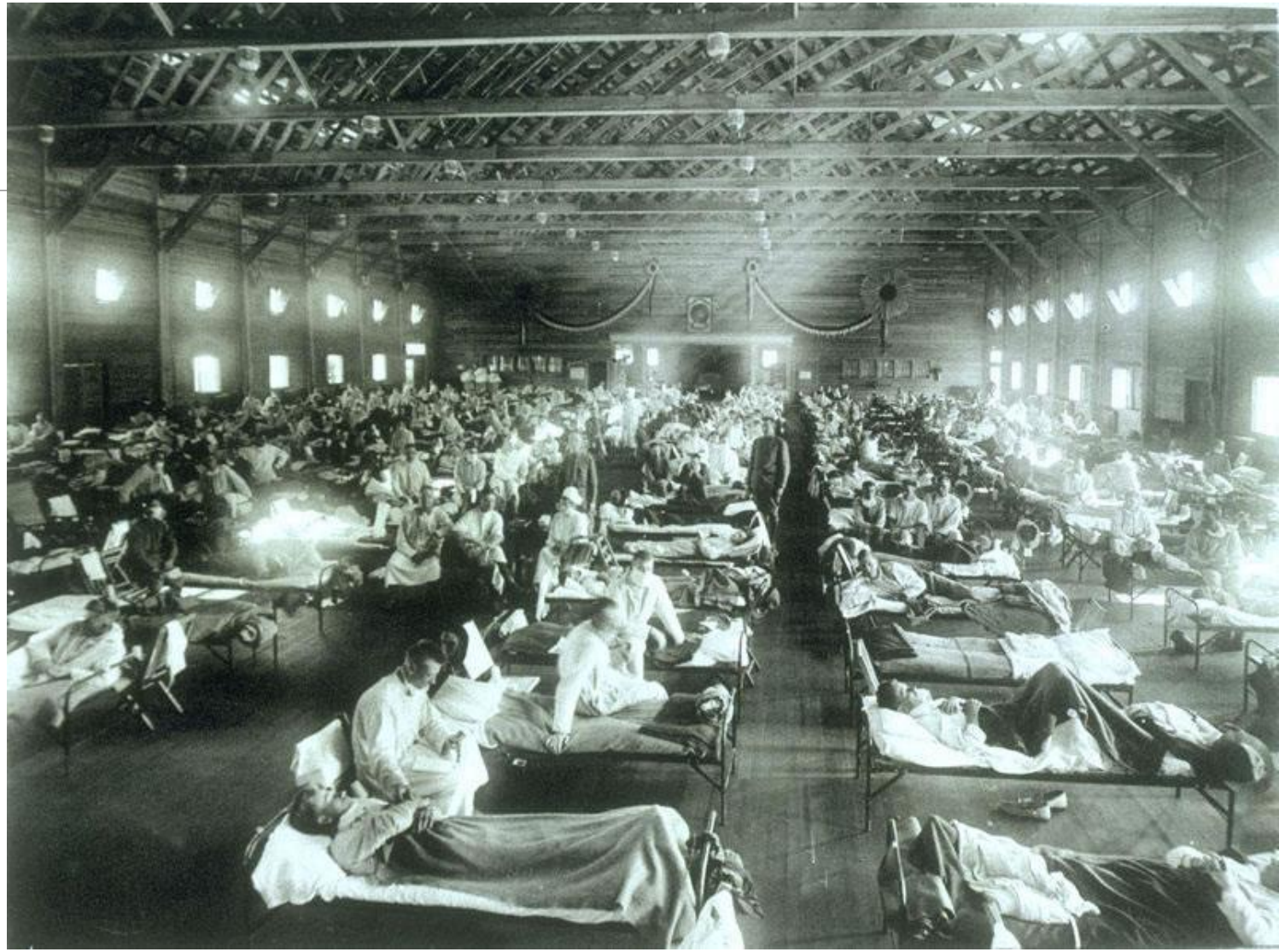


In 1890, an influenza pandemic swept the globe, killing many in its wake. [Credit: National Library of Medicine]



L'ÉPIDÉMIE D'INFLUENZA. — Vue intérieure de la tente-hôpital.

The most famous strain was the Spanish Flu, which was estimated to have killed 2-5 percent of the human population in 1918-1919.

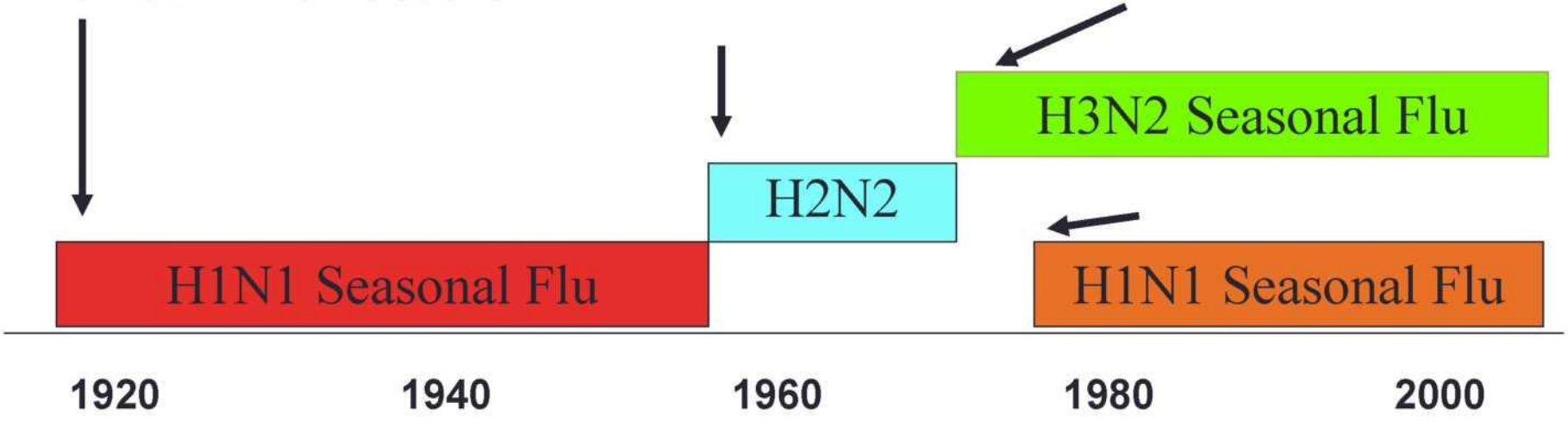


Circulating Seasonal Influenza A Sub-Types from Pandemics of the 20th Century

1918/19
40-100 million deaths

1957/58
~2 million deaths

1968/69
~1 million deaths



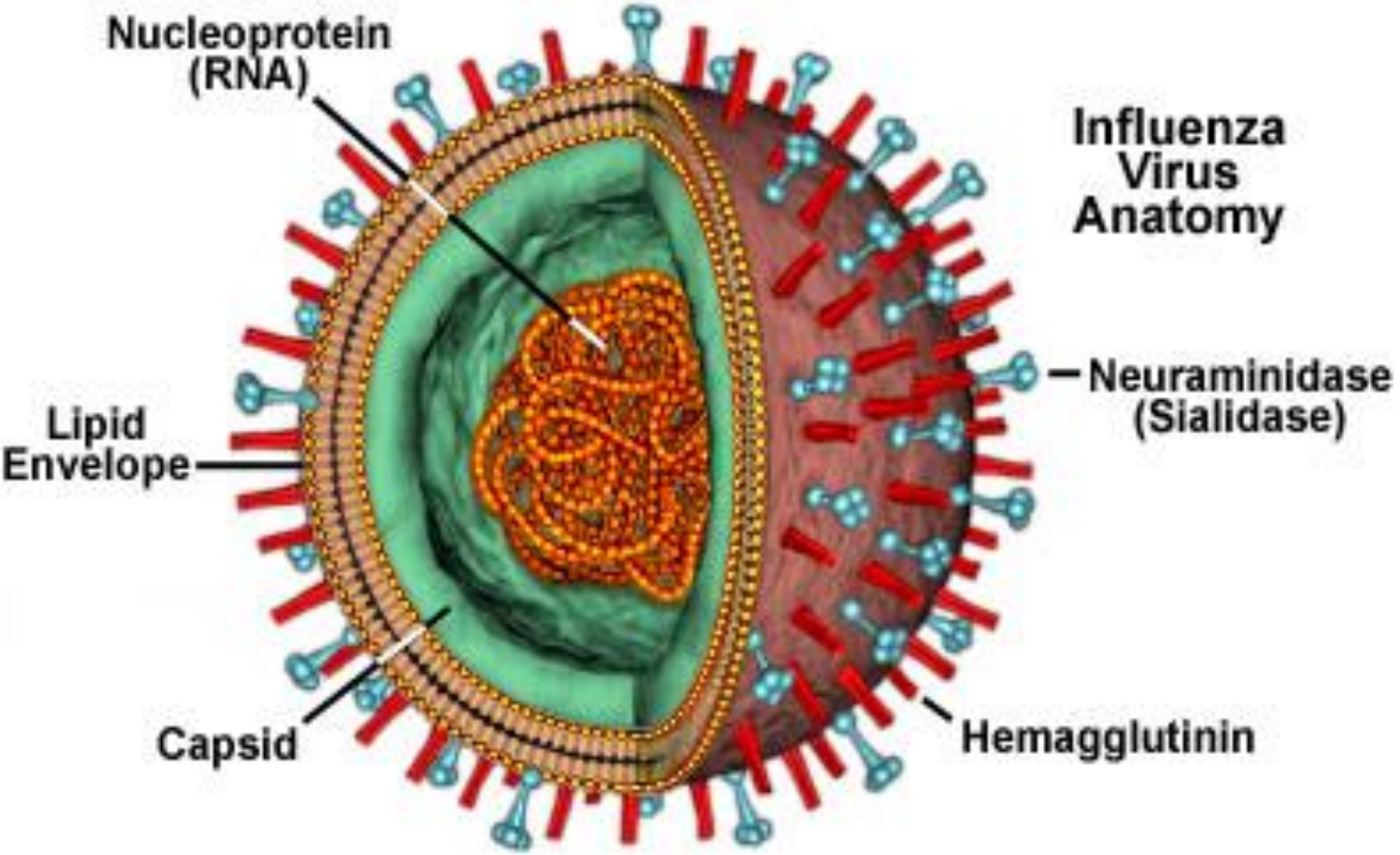
4 pandemics since 1889, with 11 to 39 years (average ~30 years) between each = ~3.3% annual risk of pandemic onset (but likely higher now)

Classification. Morphology.

Influenza virus belong to RNA-containing viruses, family Orthomyxoviruses.

The influenza viruses are spherical and measure 80-120 nm in size. The nucleocapsid is formed of a ribonucleoprotein helix enclosed in an outer lipid-carbohydrate-protein membrane.

The viral proteins consist of seven different polypeptides. Four of them are joined to the nucleocapsid and three to the outer membrane (three main antigens).


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

Antigenic structure

"S" or soluble antigen: the protein in the ribonucleoprotein core of the virus particle. All influenza A viruses share a common "S" antigen which is different from that present in all influenza B viruses; demonstrated by complement fixation test.

Haemagglutinin: contained in the radially-projecting spikes in the virus envelope; strains-specific: the main neutralizing antigen responsible for immunity to the virus. Human influenza viruses have four haemagglutinin subtypes (HO, H1, H2, H3)

Neuraminidase: also antigenic and contained in the virus envelope; plays a minor role in immunity to reinfection. Human influenza viruses have two neuraminidase subtypes (N1 and N2).

Types of virus

Based on differences of antigenic structure of the internal or nucleocapsid proteins, influenza viruses are divided into three types: A, B and C.

A: the principal cause of epidemic influenza. Influenza A viruses are also found in animals – notably birds, pigs and horses.

B: usually a milder disease but also causes winter outbreaks (especially in children)

C: doubtful pathogenicity for humans

Influenza A viruses are uniquely able to undergo frequent antigenic change. Antigenic change may be:

- ***Major:*** **antigenic shift**.
- ***Minor:*** **antigenic drift**.

Influenza B also shows antigenic variation, but the changes are less than in the case of influenza A.

Resistance *of* Virus

- Inactivated by heating at 50^0C for 30 min
- Survive at 0 - 4^0c for 1 week
- Virus preserved at - 70^0c
- Survive in the blankets for 2 weeks
- Ether, formaldehyde, Phenol destroy the virus

TRANSMISSION

AEROSOL

100,000 TO 1,000,000 VIRIONS PER DROPLET

18-72 HR INCUBATION

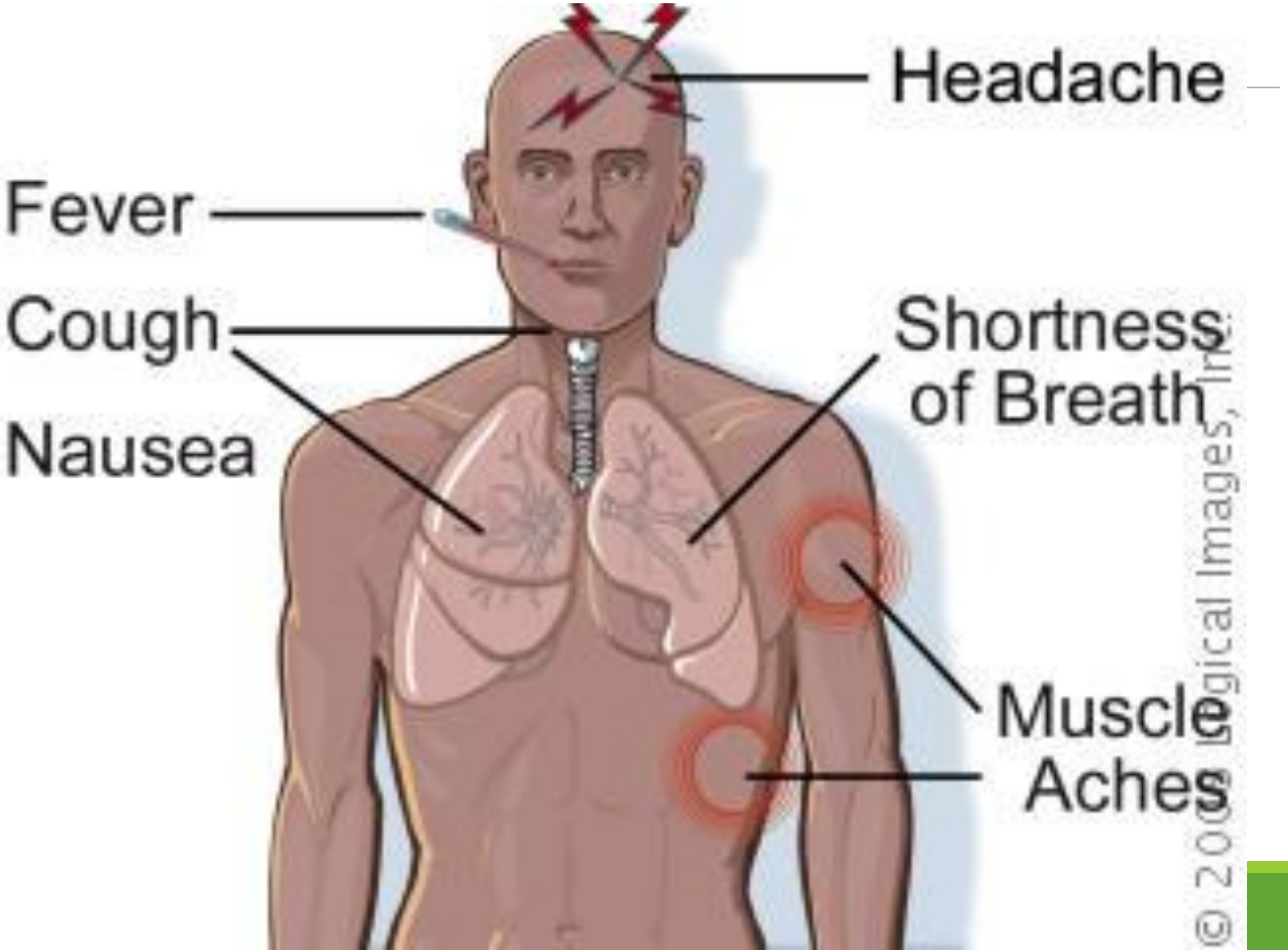
Clinical features

Influenza virus enters the body by respiratory tract. The main susceptible cells are ciliated cells of the respiratory tract whose membranes contain the specific mucoprotein receptors. Viral neuraminidase lowers viscosity of mucous. Then large number of cells are infected and killed.

The incubation period varies from 1 to 4 days. During acute illness, extensive desquamation of respiratory epithelium occurs due to necrosis of virally infected cells.

A typical case of influenza is of abrupt onset. Symptoms: fever, chills, headaches, dry cough and generalized myalgia. Fever lasts about 3 days and respiratory symptoms last for another 3 to 4 days. An uncomplicated case usually resolves within 7 days. In some cases bronchitis, pneumonia, encephalitis, influenzal meningitis.

Clinical features



CLINICAL FINDINGS

SEVERITY

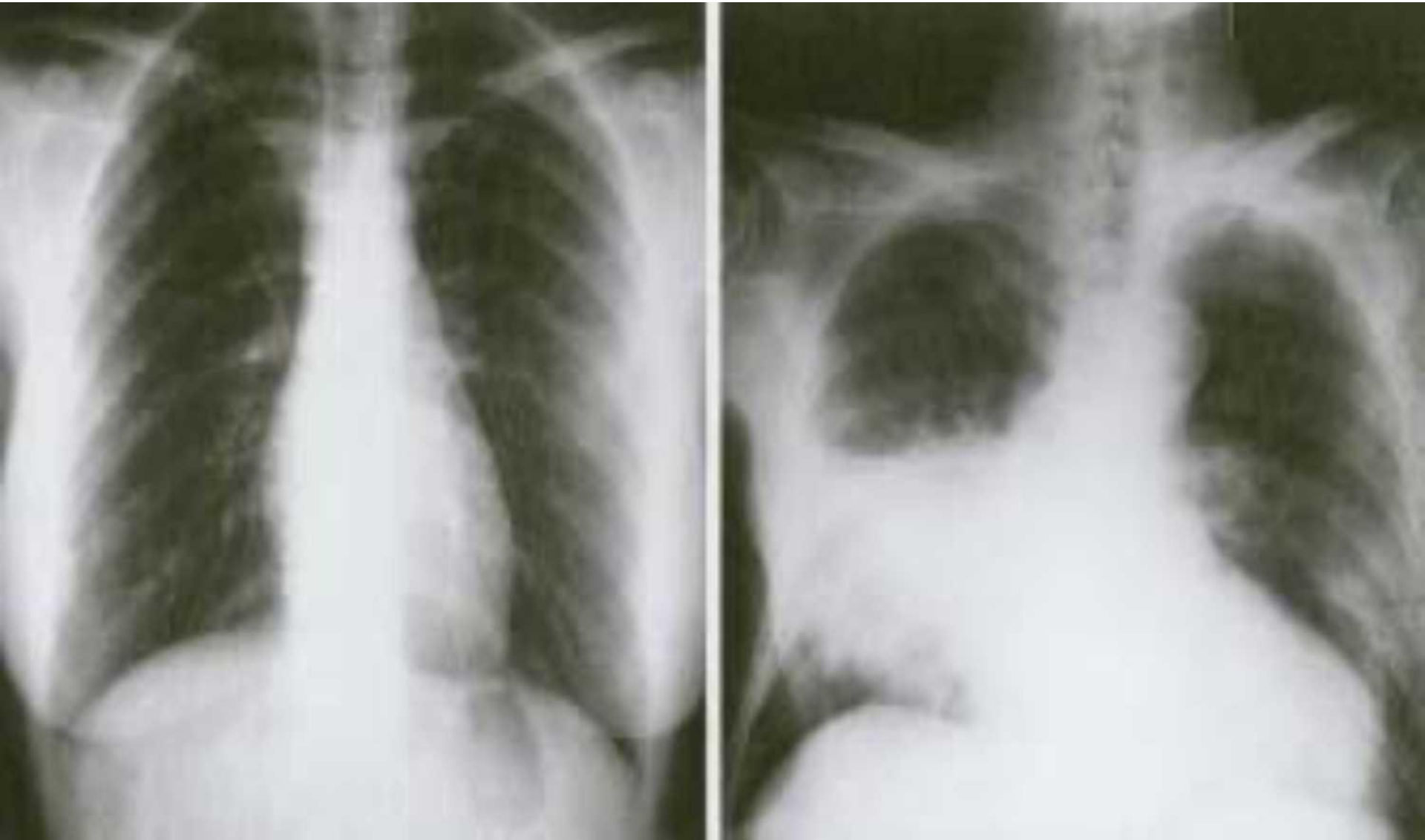
VERY YOUNG

ELDERLY

IMMUNOCOMPROMISED

HEART OR LUNG DISEASE

Viral Pneumonia is Leading cause of Death



Immunity

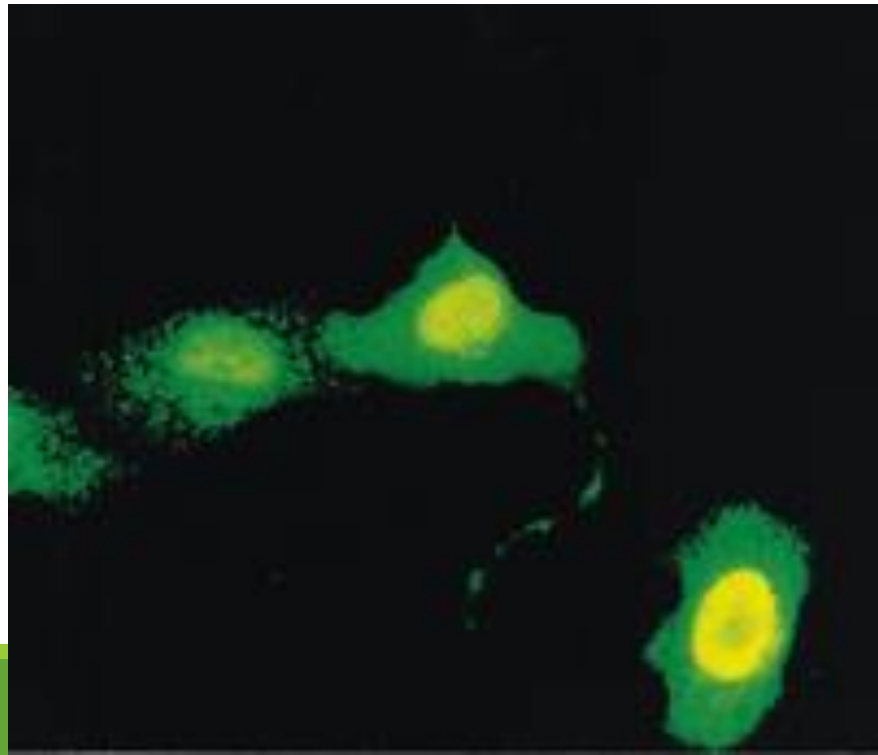
An attack of influenza produces active immunity against the strain. Protection correlates with the concentration of serum antibodies and IgA antibodies in nasal secretions. Local secretion of IgA plays the dominant role. The beneficial effect lasts for 1 to 2 years. The short duration of immunity is related to antigenic variation undergone by the virus frequently.

Laboratory diagnosis of influenza

Direct demonstration:

Specimen: nasopharyngeal aspirate.

Detect virus antigen by indirect immunofluorescence: a very rapid method of diagnosis.



Influenza A detection in infected specimen by indirect immunofluorescence method

Serology: (widely used):

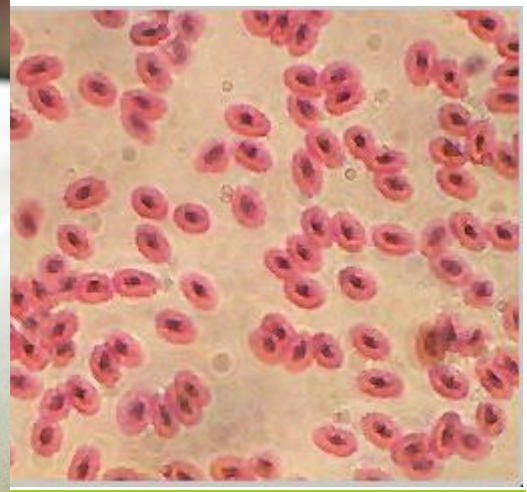
Complement fixation test: with the “S” or soluble nucleoprotein antigen.

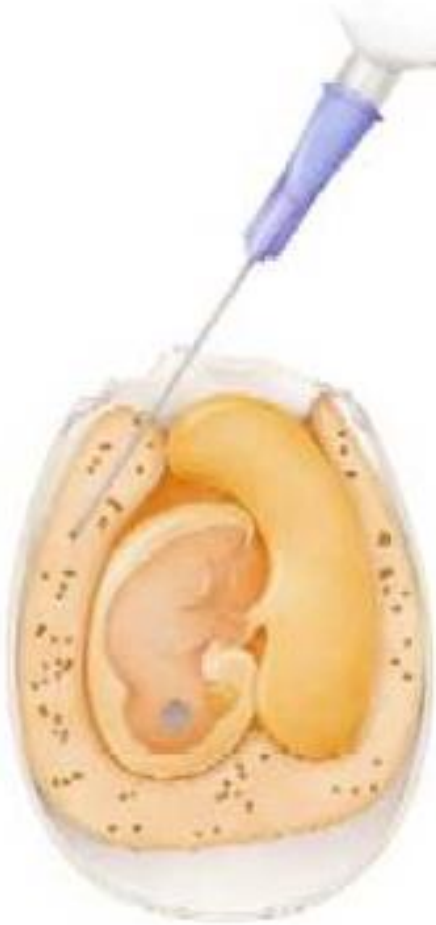
Isolation and amplification of

influenza viruses on embryonic eggs

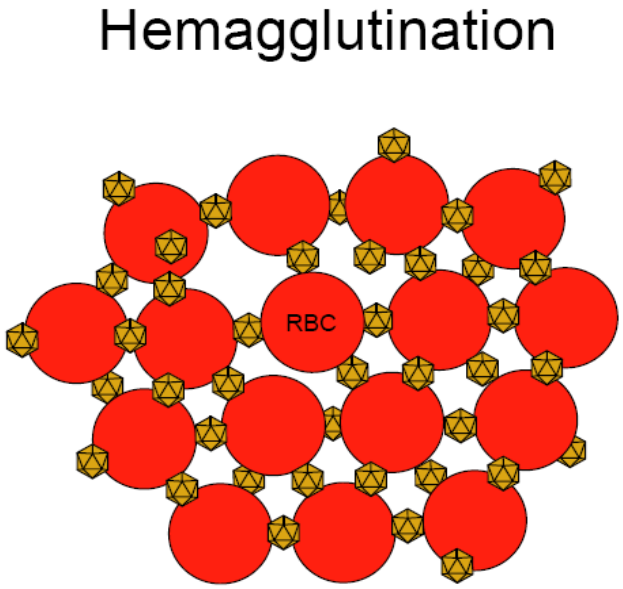
*and in cell culture (*monkey kidney

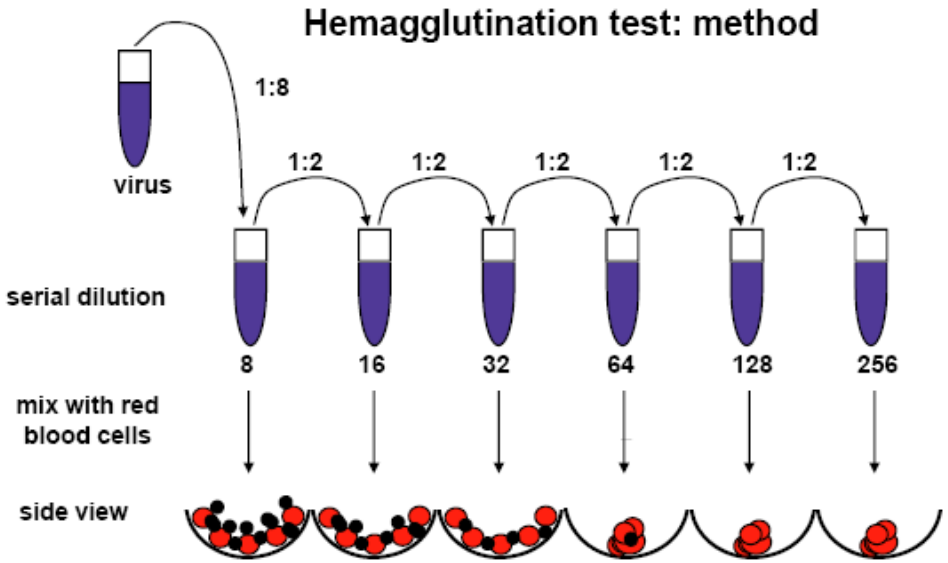
tissue cultures).





Indication (observe): for haemagglutination with human group O erythrocytes.



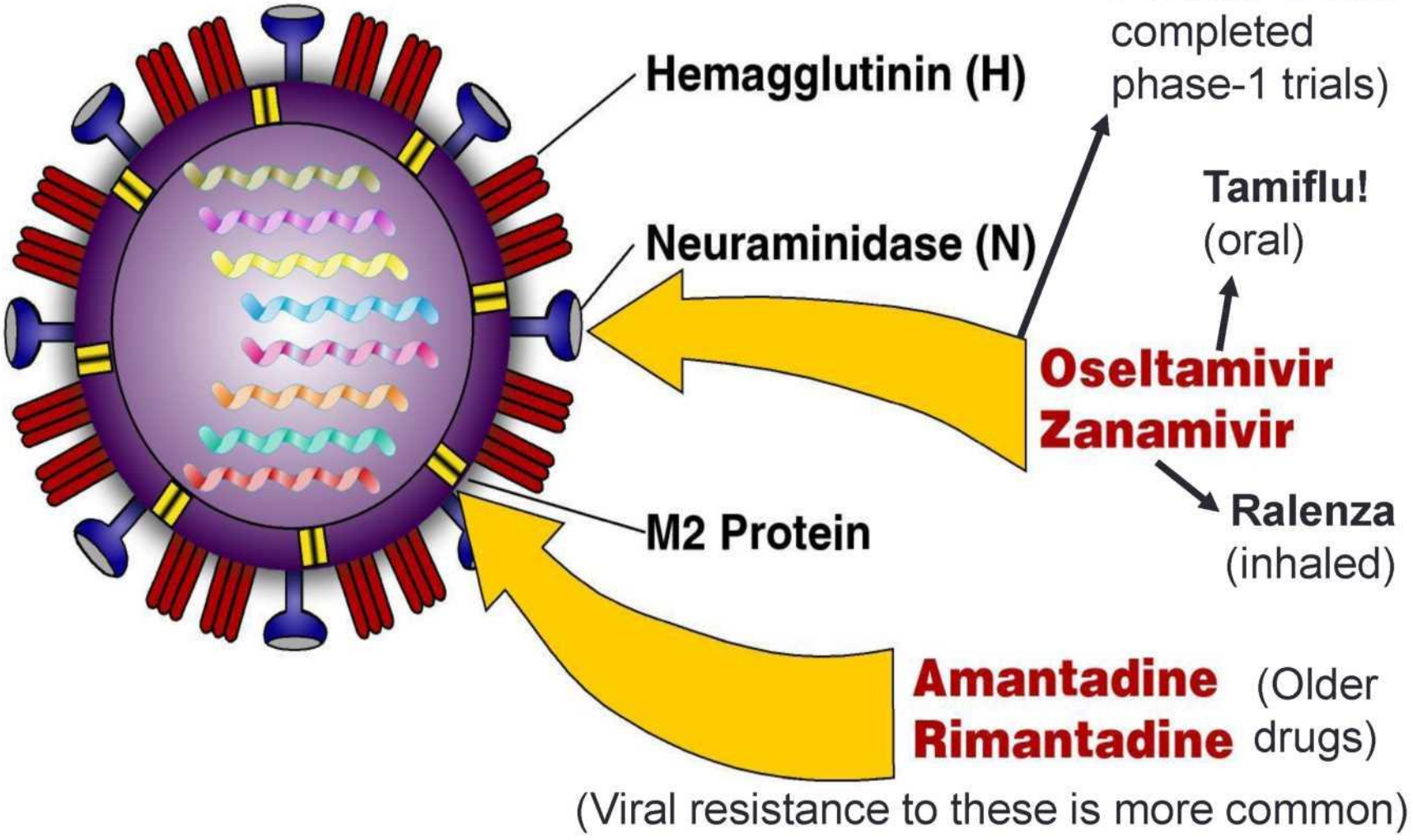


Identification (typing of virus): by complement fixation: strain identification by haemagglutination-inhibition in a reference laboratory

Treatment

As with many other viral diseases, only the symptoms of Influenza usually are treated. However, the antiviral drugs **Amantadine (Symmetrel), rimantadine (Flumadine), zanamivir (Relenza), and oseltamivir (Tamiflu)** have been shown to reduce the duration and symptoms of type A influenza if administered during the first two days of illness. All four of these drugs attack the virus directly by plugging the catalytic site of the enzyme neuramidase. With the enzyme inactivated, viral particles can't travel from cell to cell. Interferon (inhibits virus replication).

Aspirin (salicylic acid) should be avoided in children younger than 14 years to reduce the risk of Reye's syndrome.


(Injectable
Peramivir has
completed
phase-1 trials)
Hemagglutinin (H)
Neuraminidase (N)
Tamiflu!
(oral)
Oseltamivir
Zanamivir
Ralenza
(inhaled)
M2 Protein
Amantadine
Rimantadine
(Older
drugs)
(Viral resistance to these is more common)

Vaccination.

Vaccination is currently recommended for those suffering from cardiac or respiratory problems especially if elderly. This is to protect those at high risk of death or serious complications.

Vaccination is not aimed at preventing transmission, and can not do so.

Virus vaccine

Contains: inactivated virus grown in chick embryos: either purified subunits (i.e. surface antigen) or disrupted virus purified and ether-treated to solubilize envelope proteins.

Current vaccine: contains three strains: influenza A, H1N1 and H3N2, influenza B.

Administration: intramuscularly, annually.

Protection: relatively short-lived (around a few months): effective but not solid immunity (around 60% protection) conferred.

Live attenuated virus vaccines

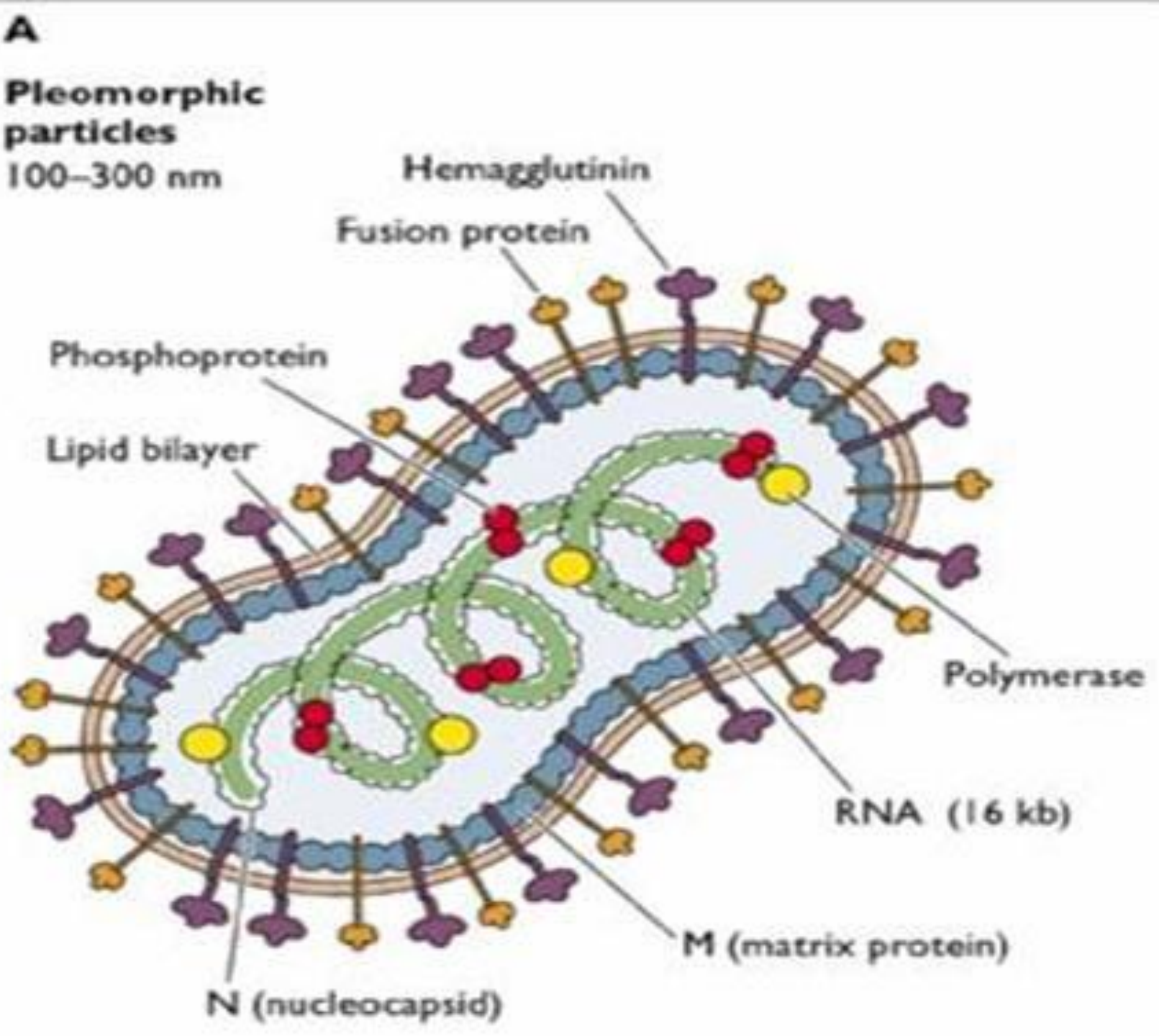
Administration: intranasally: not yet generally accepted.

Measles

Classification. Measles virus belong to the family Paramyxoviridae, genus Morbilli viruses.

Morphology. The measles virus is spherical shape. Diameter of virion is 120-250 nm. Its core is a single negative strand of non-segmented RNA, associated with proteins (P and NP) and an RNA polymerase.

The outer membrane consists of inner matrix protein (M) and the envelope lipid bilayer, covered by projections of 12-14 nm x 2-4 nm. Spikes on envelope contain a haemagglutinin but no neuraminidase. It also contains an F protein.


A
Pleomorphic particles
100–300 nm
Hemagglutinin
Fusion protein
Phosphoprotein
Lipid bilayer
Polymerase
RNA (16 kb)
M (matrix protein)
N (nucleocapsid)

Replication. Measles viruses attach via the haemagglutinin glycoprotein to the sialic acid-containing receptors on host cell. The virus penetrates the cell by means of fusion of viral and cell membranes, which is mediated by F4 glycoprotein. F4 glycoprotein becomes activated only when it is cleaved into two subunits, F41 and F42. If F4 protein fails to cleave, the virus will attach but will not fuse with the host cell membrane and the viral genome cannot penetrate the cell. The negative strand genome cannot act as mRNA, the viral RNA polymerase transcribes mRNA in the cell cytoplasm. Viral components are synthesised in the cytoplasm. The surface H and F proteins are incorporated into a stretch of membrane forming a viral envelope that encloses the virus. The enveloped virus buds out of the cell into the exterior.

Epidemiology

Man is the only natural host of measles. Transmission occurs predominantly by respiratory route, conjunctivitis may also be a source. Infection is transmitted by the patient during few days before and after the rash. The disease is endemic throughout the world and epidemics occur in late winter and early spring.

Pathogenesis and clinical features

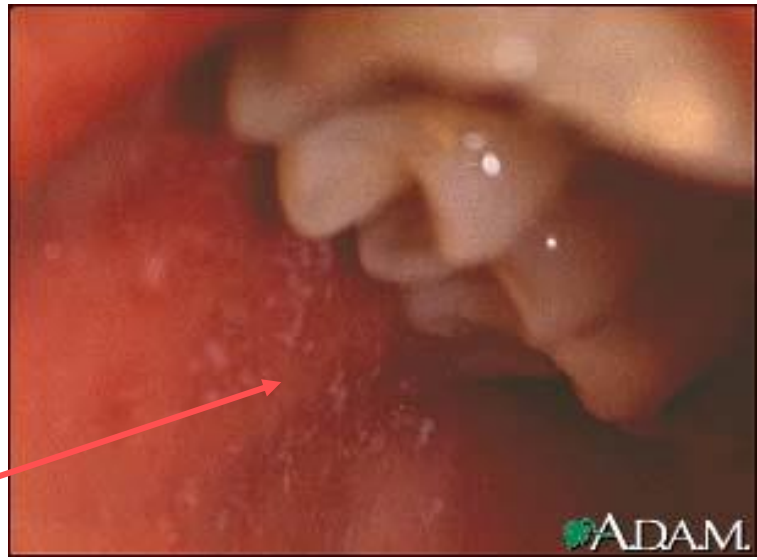
Measles virus is acquired by inhalation. The incubation period is 9-12 days.

Virus multiplies in lymphoid tissues of respiratory tract and invades blood stream, affecting endothelium.

The earliest features associated with phase of viraemia consist of high fever, cough and conjunctivitis.

In this prodromal phase,

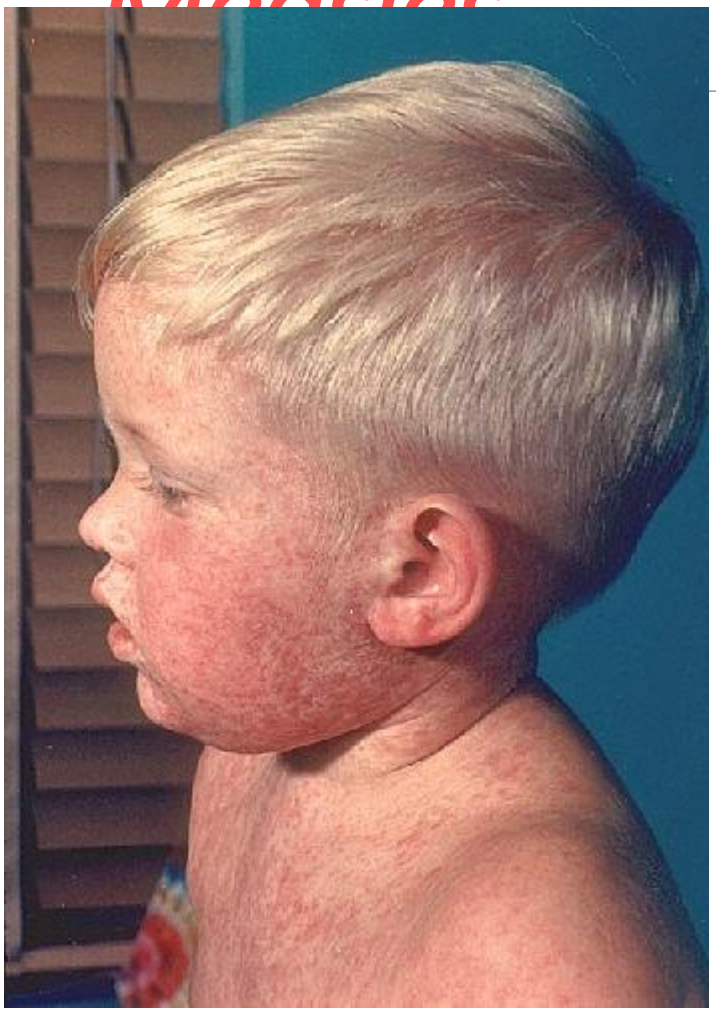
Koplik`s spots (red spots with a bluish white centre on the buccal mucosa) may be present.

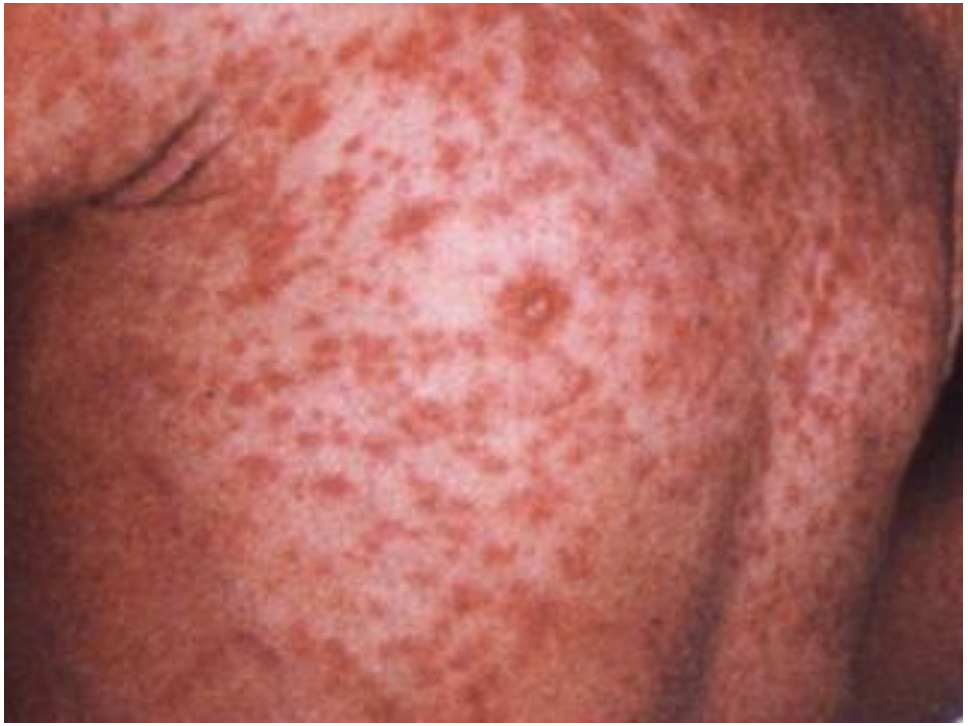


Koplik`s spots

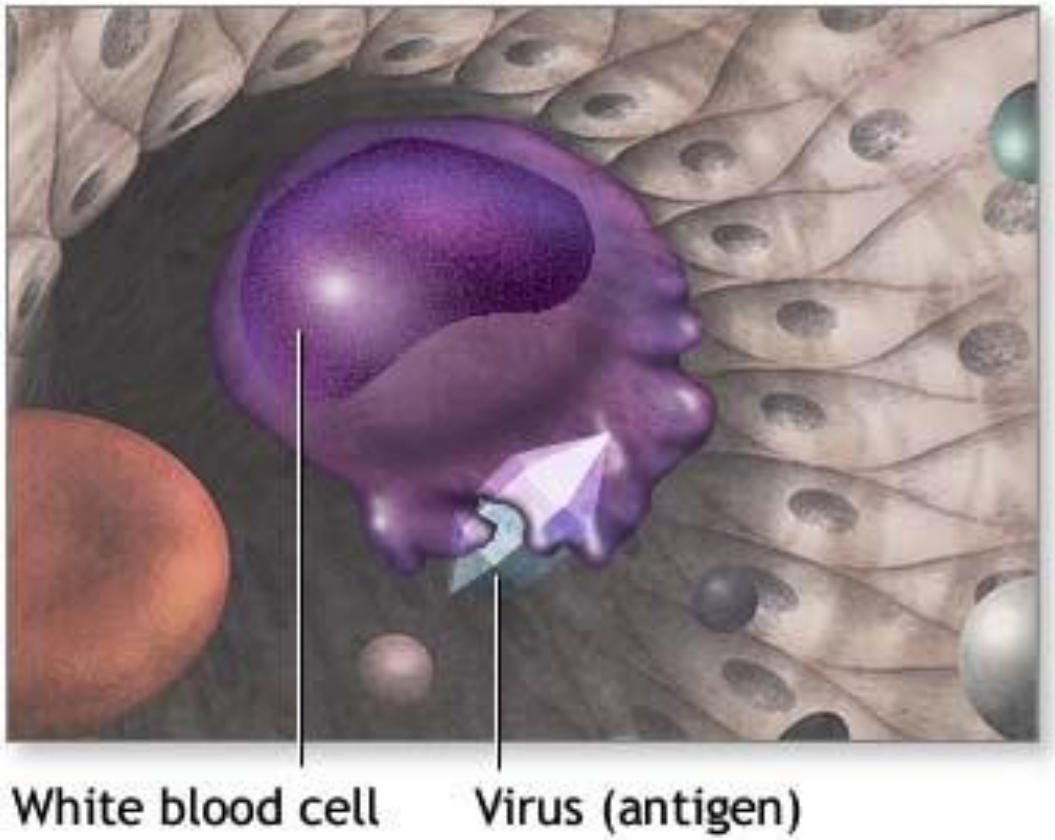
From blood, the virus localises in epithelial surfaces of skin, respiratory tract and conjunctiva. With the decline of acute symptoms in 1-2 days, widespread-maculopapular rash appears on skin, mucous membranes and conjunctiva. Rash results from interaction of immune T cells with virus infected cells in small blood vessels. The rash fades in about a week and most patients recover by 10-14 days. Complications of measles may be pneumania, otitis media and a subacute sclerosing panencephalitis (SSPE). SSPE is a slow form of measles virus infection with lethal outcome. In SSPE the defective form of measles virus persists within the infected cell. Because of its inability to induce production of a functional M proteins, it is not released as complete virus from the cells. Patient`s condition deteriorates over several years and finally develop CNS symptoms: dyskinesia, mental deficiency.

Measles





Immunity. There is only one serotype of measles virus and one infection confers life-long immunity.



Laboratory diagnosis

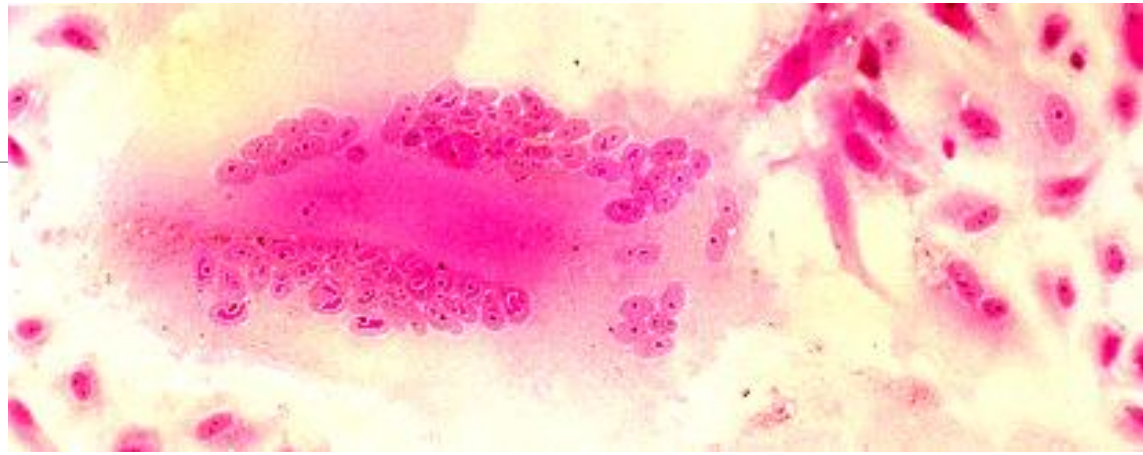
Immunofluorescent study of exfoliated respiratory cells in nasopharyngal secretions show virus particles. Smears stained by Giemsa`s stain show giant cells and inclusion bodies.

EIA with application of anti-IgM serum is used.

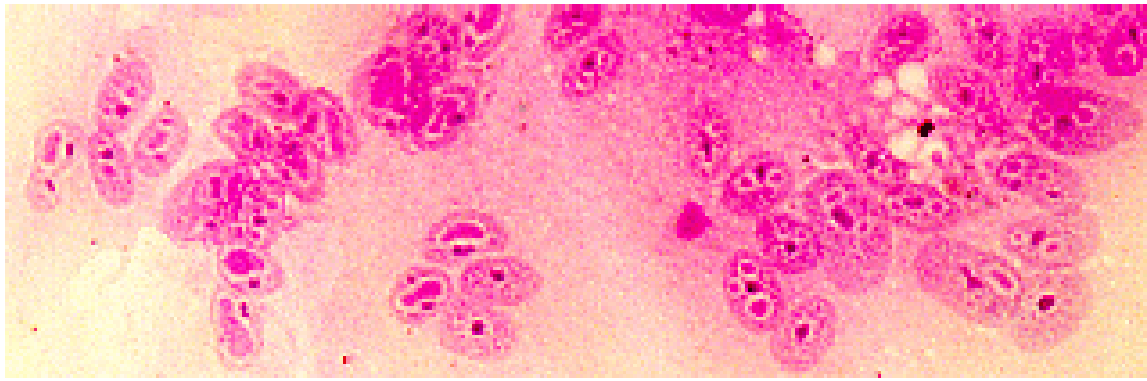
Isolation of virus: Specimens (nasopharyngal swab, blood sample) collected during febrile period are cultured in human fibroblasts, monkey or human kidney cells. Growth occurs slowly with CPE (multinucleated cells) containing both intranuclear and intracytoplasmic inclusion bodies in 7-10 days.

Serology: Pising titre of measles-specific IgM antibodies can be detected by HAIT, CFT and NtT.

Very large syncytia can be formed during replication of measles virus in cell culture.



*In the **syncytium** shown on the right, multiple nuclei are clustered around an eosinophilic cytoplasmic mass that probably represents the Golgi compartments of the fused cells.*



Intra-nuclear inclusions are clearly visible.

Prophylaxis

Active immunization: Live attenuated vaccine is 90% effective. Measles vaccine is being used in combination with mumps and rubella (MMR Vaccine). The vaccine is administered at 12 to 18 months of age, after complete disappearance of maternal antibodies.

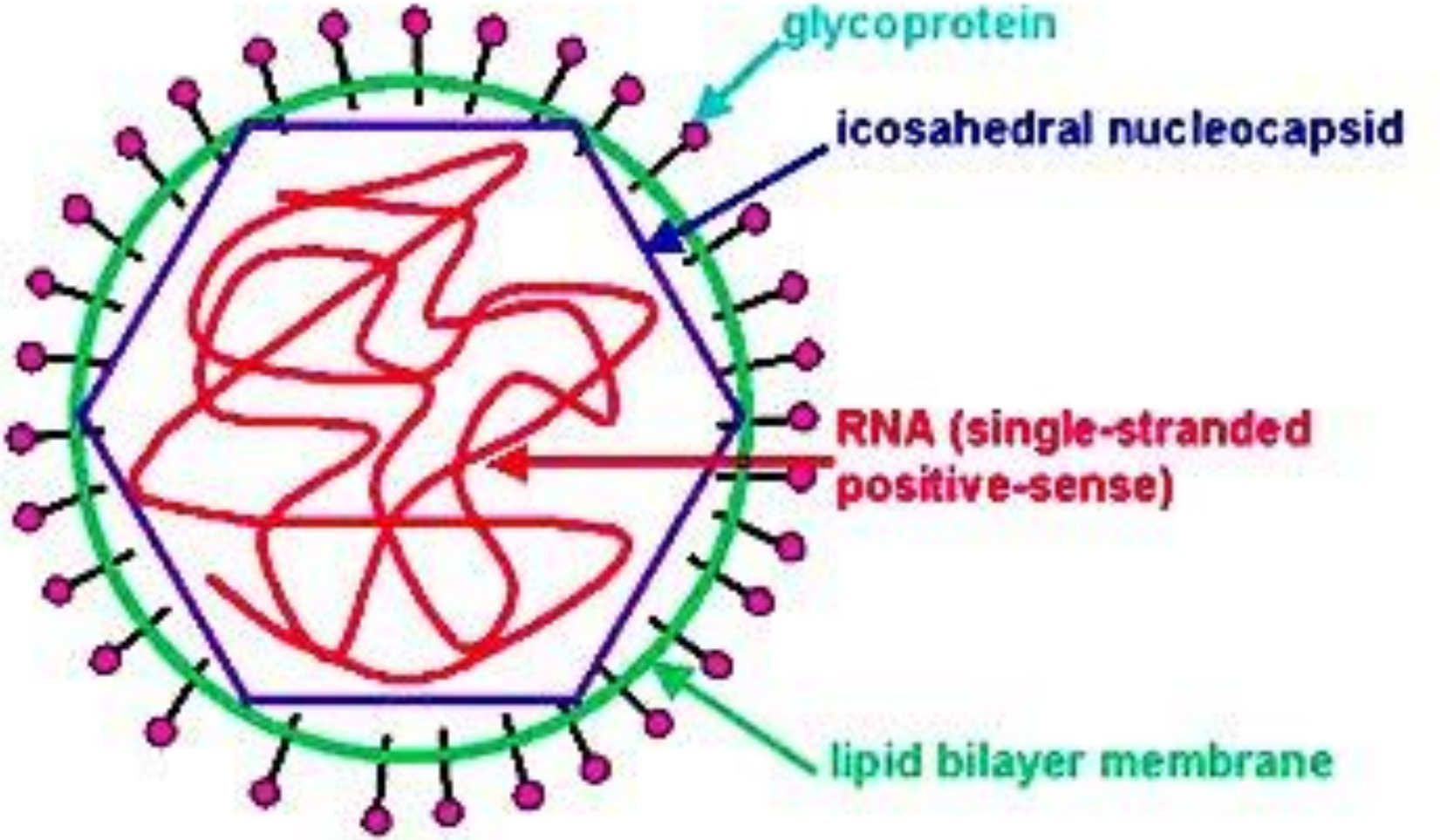
Passive immunization: Anti-measles immunoglobulin serum.

Rubella

Classification. Rubella or German measles virus is a representative of Togaviridae family and a sole member of the Rubivirus genus.

Morphology. The rubella virus is pleomorphic with diameter of virion particle about 50-70 nm. Its core is a single positive strand of RNA. Nucleocapsid has icosahedral symmetry and diameter about 30 nm. The enveloping membrane consists of proteins and lipids. The outer surface of membrane is covered by projections,containing a haemagglutinin.

RUBELLA VIRUS



Epidemiology

Rubella is worldwide in distribution. Before the vaccine was introduced, over 50,000 cases of rubella per year were reported, but this incidence is much lower than the true frequency of the disease, which is mild, often subclinical, and difficult to diagnose. Most infections occur during the late winter and early spring months. Epidemics occur every 5-6 years.

Pathogenesis and clinical features. Rubella may be acquired congenitally or post-natally

Postnatal rubella

Transmission is from person-to-person through inhalation of infected aerosolized respiratory secretions. The incubation period is 12-23 (18 days on average). The rubella virus penetrates through the mucosa of upper respiratory tract and multiplies locally in the cervical lymphoid nodes. In week virus particles invade blood stream. The feature associated with phase of viraemia is a mild fever. The viruses are then seeded into target organs from blood. It coincides with appearance of rash, accompanied by retroauricular lymphadenopathy (usually bilateral) and joint pain (especially in girls). Macular rash first appears on the face and then spreads to the trunk and legs and rarely lasts for more than 3 days. Complications: arthritis, thrombocytopenia and hemorrhagic diathesis, postinfectious encephalitis.

Congenital rubella

During viraemic phase, rubella virus is able to cross the placental barrier and can replicate in differentiating cells of the embryo. Supression of mitotic activity of foetal cells and affection of placental vessels are the causes of teratogenic effect. The extent of teratogenic effect depends on the timing of foetal infection. The risk is maximum in the first trimester of pregnancy (50-70%), followed by 2nd trimester (20 %) and only 4% in 7th month of pregnancy.

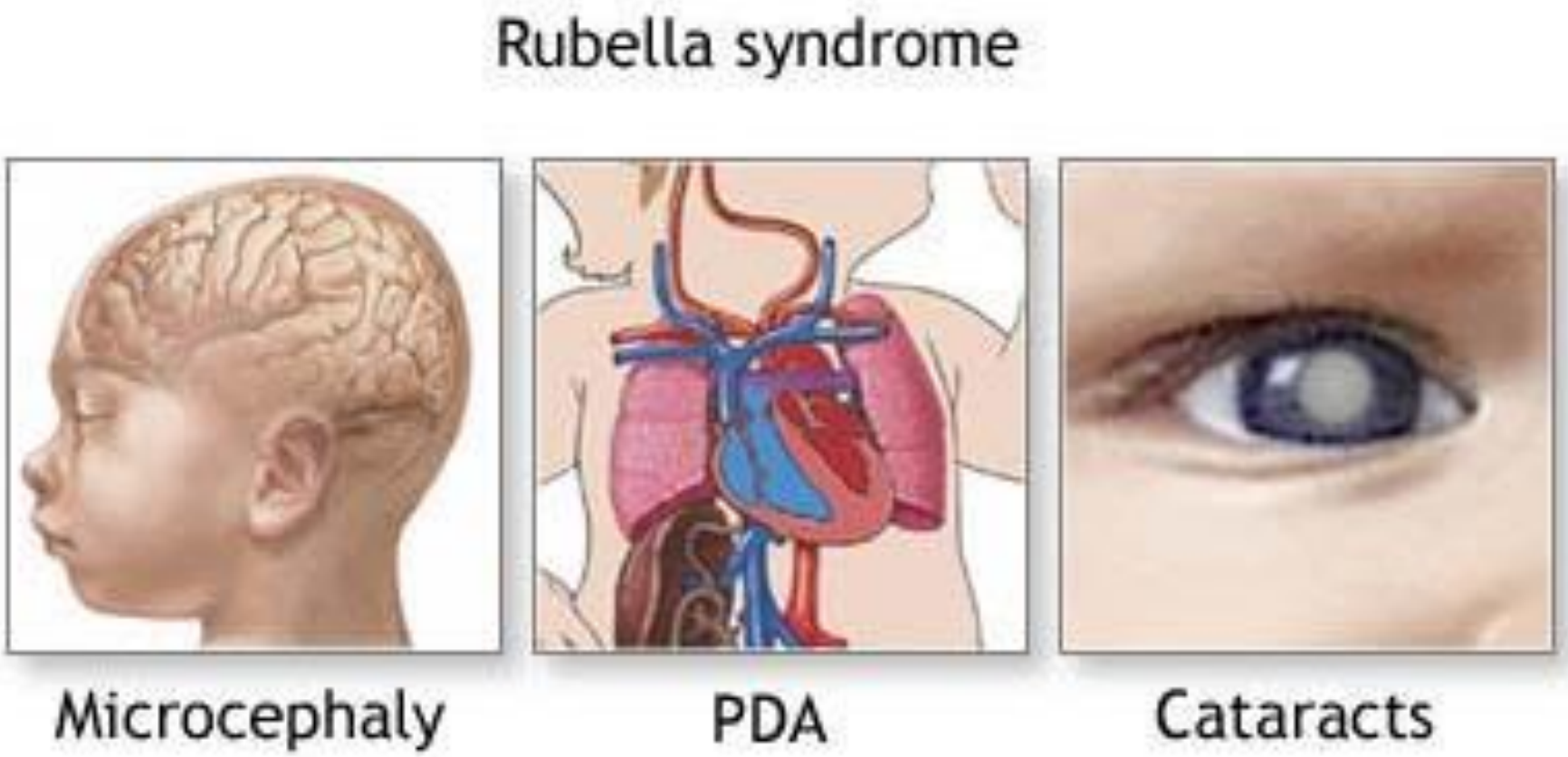
Fetal defects can be transient or permanent (the latter are usually the result of developmental abnormalities).

Transient abnormalities include low birth weight, thrombocytopenic purpura, hepatosplenomegaly.

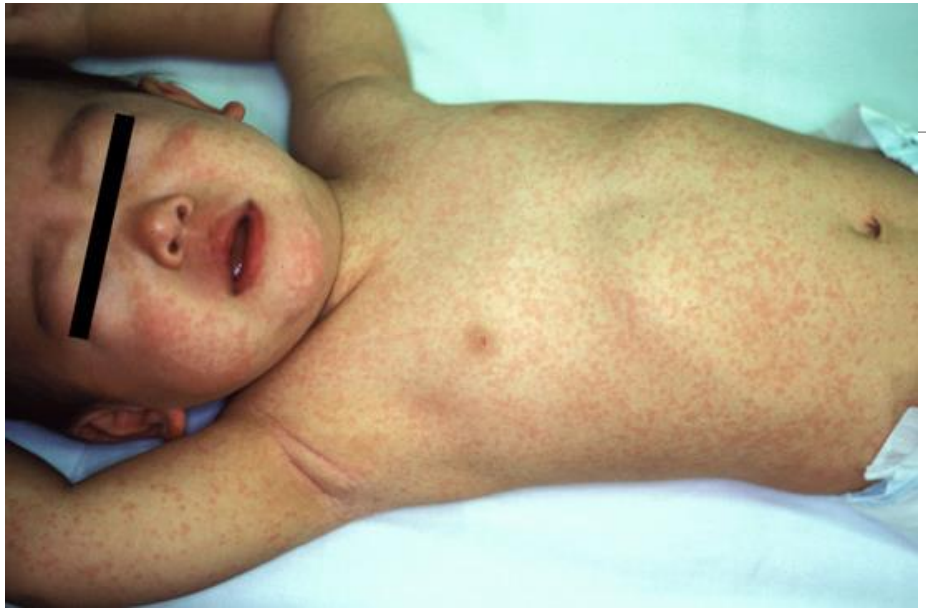
Permanent abnormalities include cataract, cardiac lesions, microcephaly and deafness (the classical congenital rubella syndrome).

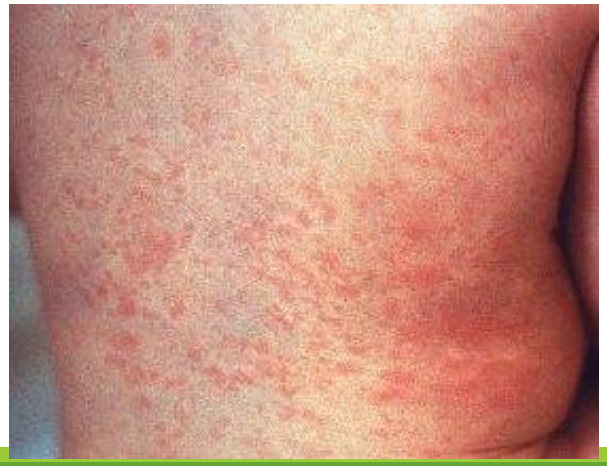
***Immunity*.** There is only one serotype of rubella virus and one infection confers life-long immunity. Immune mothers transmit antibodies to breast-fed infants for 4-6 months.

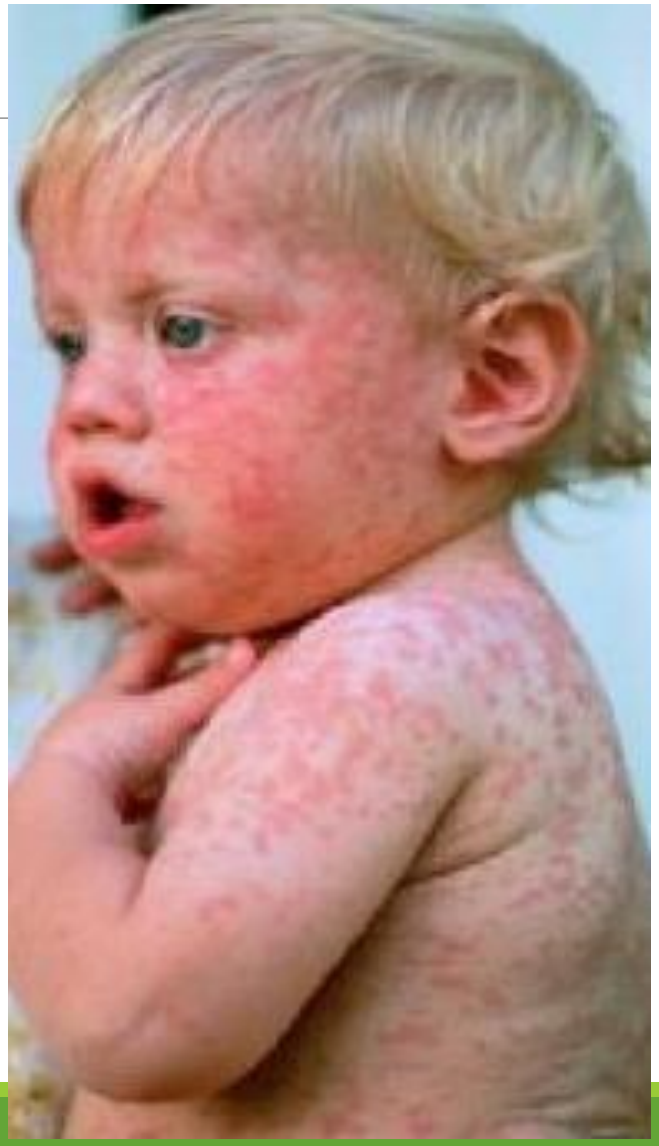
Congenital Rubella Syndrome (CRS)



This is a good example of a maculopapular rash.







Laboratory diagnosis

Rapid methods of diagnosis are absent. The diagnosis is established by isolation of the virus from clinical materials, or, more commonly, by serology.

Detection of virus

Material: Blood, throat swabs (during acute phase of disease); blood, urine, stool (after appearance of rash)

Cultivation in cell culture (PK-13, SIRC, BSC-1, Vero). CPE is inconspicuous.

Identification of virus

1. HAIT
2. Interference technique (in which inoculated monkey kidney cells are challenged by Coxsackievirus A 9).

Detection of antibodies

IgM antibody: recent infection is best diagnosed by detection of IgM by ELISA or immunofluorescence.

Single radial haemolysis: widely used for detecting immunity in pregnant women or in women at special risk, e.g. children's nurses, schoolteachers: it does not measure antibody titre and is less useful for the diagnosis of rubella.

Prophylaxis. Objective of vaccination is to protect unborn babies rather than the vaccine. Vaccination has been advocated in women of child-bearing age and school girls of 11-13 years age. Live, attenuated vaccine grown in human diploid cells is now widely used. The vaccine virus multiplies in the body but does not spread to contacts. Vaccine virus can cross placenta but they are not teratogenic. Rubella vaccine is being used in combination with mumps and measles (MMR Vaccine) and is given in two doses. Although the attenuated virus has not been shown to cause clinical infection or fetal abnormalities, vaccination should be withheld from pregnant women and vaccinated women should avoid pregnancy for 3 months following vaccination.

Gamma-globulin can be administered to a pregnant woman who has been exposed to rubella virus in an attempt to prevent infection. However, if the infection has occurred, gamma-globulin will not prevent viremia or fetal infection.

Thank you for attention