

LECTURE XIV

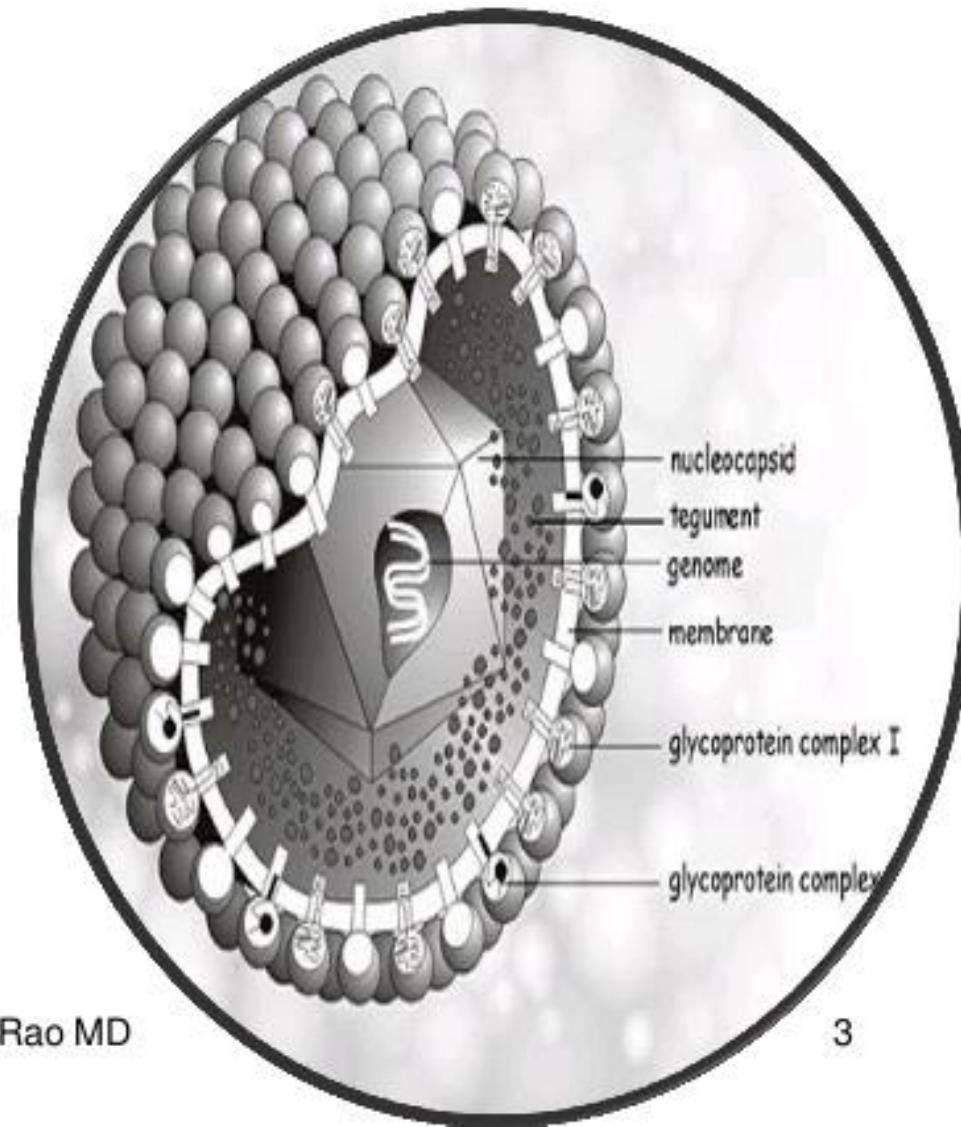
Herpes viruses, picornaviruses
and rhabdoviruses, arboviruses

Herpesviridae

- The *Herpesviridae* are a large family of DNA viruses that cause diseases in animals, including humans. The family name is derived from the Greek word *herpein* ("to creep"), referring to the latent, re-occurring infections typical of this group of viruses. *Herpesviridae* can cause latent or lytic infections.

Herpes Viruses **DNA group**

- Most important Human Pathogens
- Wide Host cell range
- Life Long Infection – Periodic reactivation
- Immunocompromised
- Large number of genes,
- Some viruses susceptible to treatment.



Infecting Humans.

Herpes Simplex virus 1 and 2

Varicella Zoster Viruses

Cytomegalovirus virus

Epstein Barr virus

Human Herpes viruses 6, 7.

**Kaposi's Sarcoma associated
Viruses**

Properties of Herpes Viruses.

- Spherical in Shape
- Icosahedral 150 to 200 nm in size
- Genome – Double stranded DNA
Linear
- Envelope contains Glycoprotein's



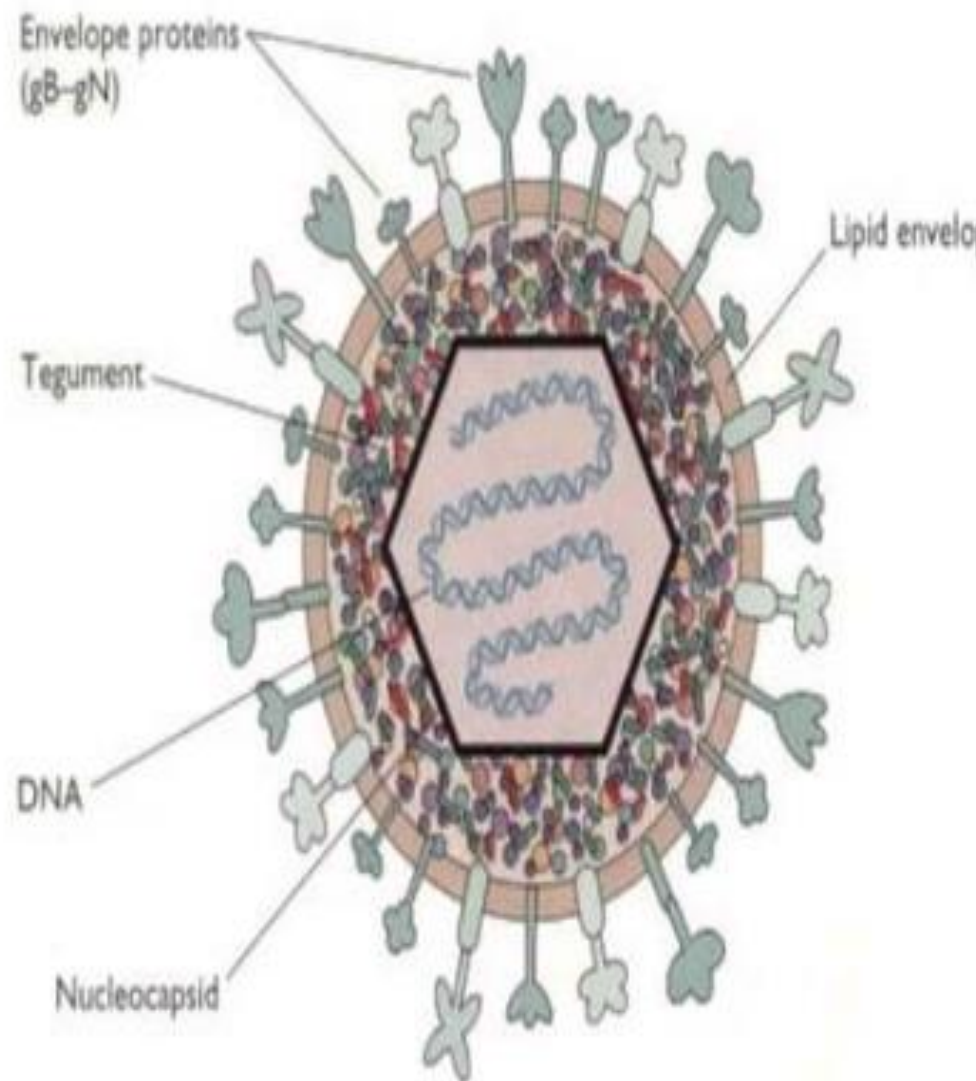
Herpes Virus Replication

- Replicates in Host Cell Nucleus
- Form Cow dry A Type inclusion bodies.
- More than 50 different types protein in infected cell.
- Large number of enzymes in DNA synthesis

Herpes Simplex 1 and 2

Herpesviridae

- Group: Group 1
Family: Herpesviridae
Subfamily: Alphaherpesvirinae
Genus: **Simplexvirus**
- **Species**
- **Herpes simplex virus 1 (HWJ-1)**
Herpes simplex virus 2 (HWJ-2)

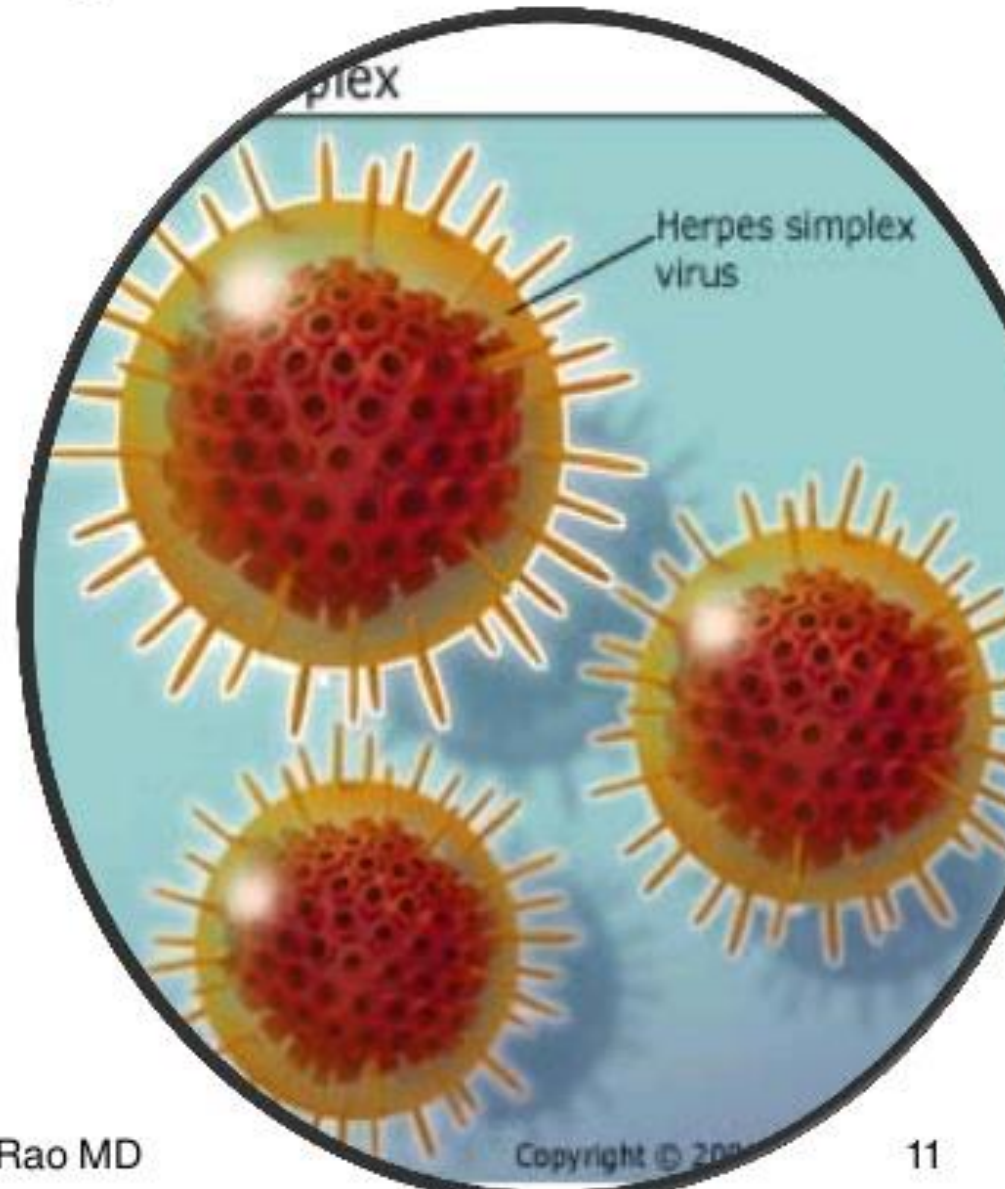


Herpes Simplex Virus

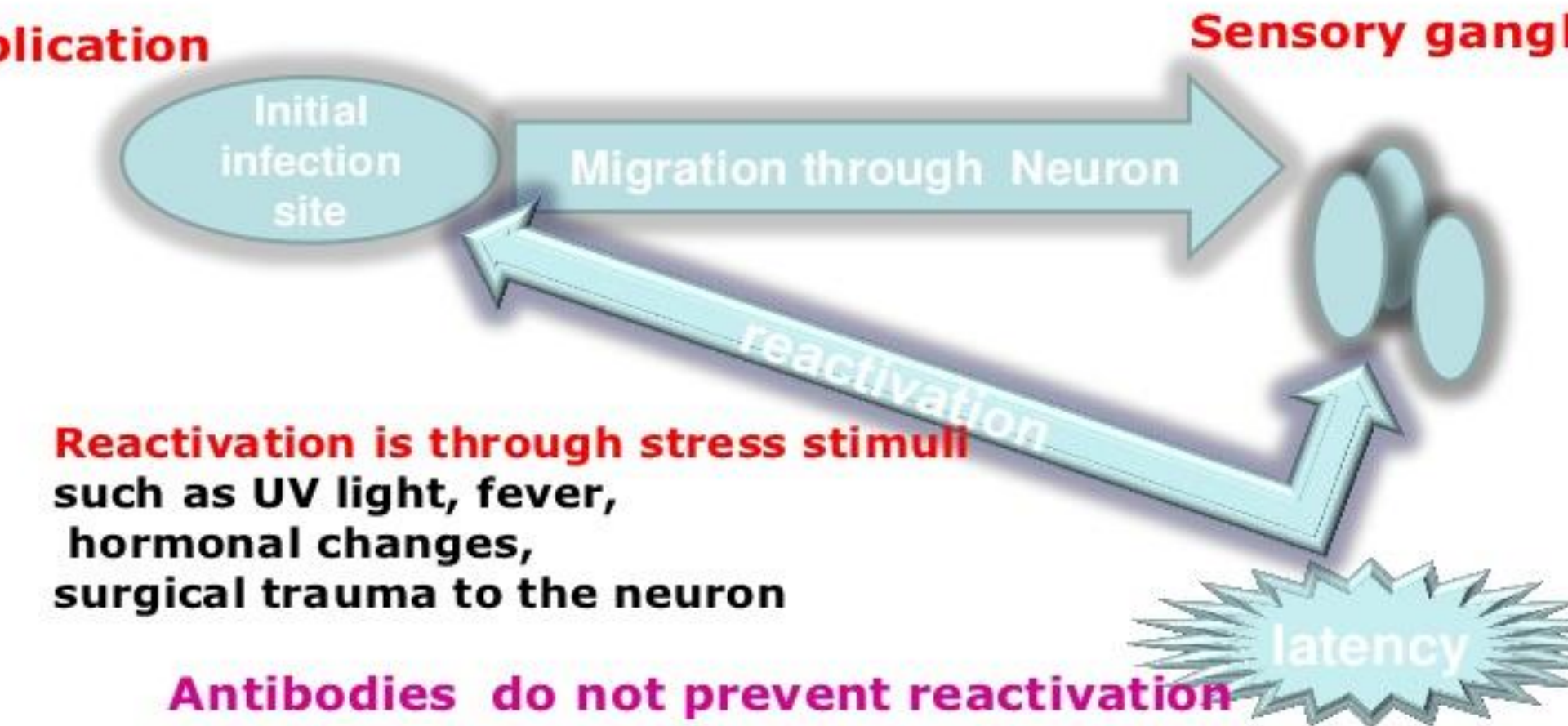
• HSV are
spherical in
shape

• Ds DNA

• 35 proteins



Pathogenesis of HSV 1 &2



HSV-1: trigeminal ganglia
HSV-2: sacral ganglia

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Infections in Humans.(Herpes Simplex 1 and 2)

- Wide spread in Humans
- Broad Host Ranges.
- Replicate in Many types of Cells.
- Produce cytolytic effects
- **Most Common Diseases.**
- Gingival stomatitis, Kerato conjunctivitis
- Encephalitis Genital diseases,
- New Born Infections, Latent Infections in Nerve Cells,
- Recurrence.

HERPES SIMPLEX VIRUS (HSV)

- **HSV 1 infect the upper part of the body**
 - **mouth and the face**
- **HSV 2 infect the lower part of the body**
 - **genital infections**
- **There is little cross protection**
- **Therefore, one can get both the**

Properties of Herpes Simplex Viruses

Type 1 and 2

- Similar in Organization
- Restriction Enzyme Differentiates
- H S V 1 contact with Saliva.
- H S V 2 Sexual
- Maternal infection (Genital Infection spreads to New Born)
- Replicates in 8-16 hours.

Virus Grows in the following.

- Primary and Continues Cell lines.
- Monkey and Rabbit Kidney,
- Human Amnion
- Syncytial formation and Giant cell formations
- Multiplies in Chorio Allontoic membrane
- Monoclonal Antibodies differentiates Type 1 and 2 types.



Fig. 11.1 Primary herpes simplex infection. Stomatitis with satellite vesicles over the chin. (Reproduced, with permission, from *Diseases of Infection* by N.R. Grist, D.O. Ho-Yeo, E. Walker and G.R. Williams, 1988. Oxford University Press.)

Pathogenesis

Entry by skin or mucous membranes

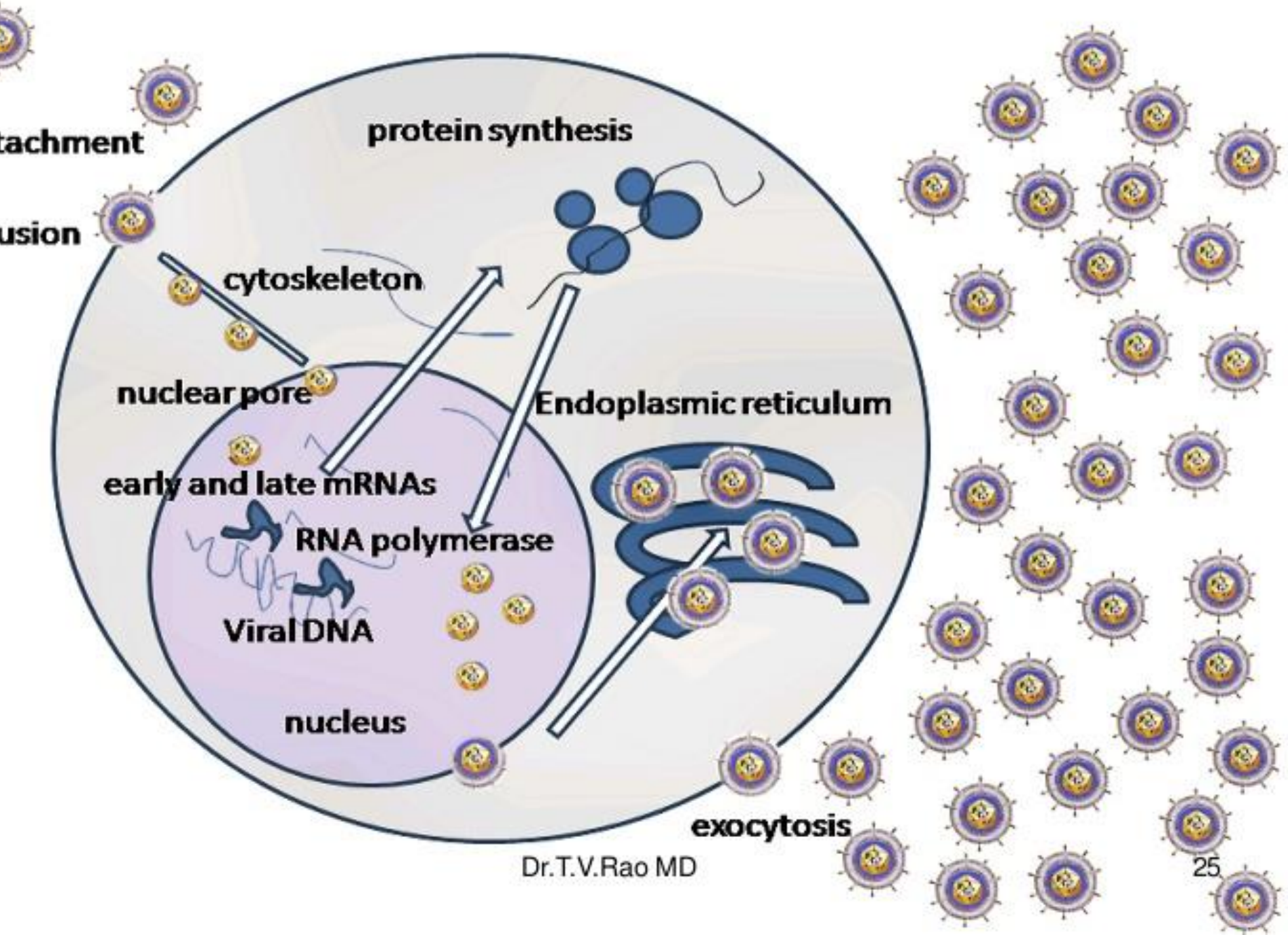
↓
Viral multiplication → sensory nerve

↓
Lysis of cells
↓
vesicles
↓
Ulcers

↓
root ganglia
↓
latency

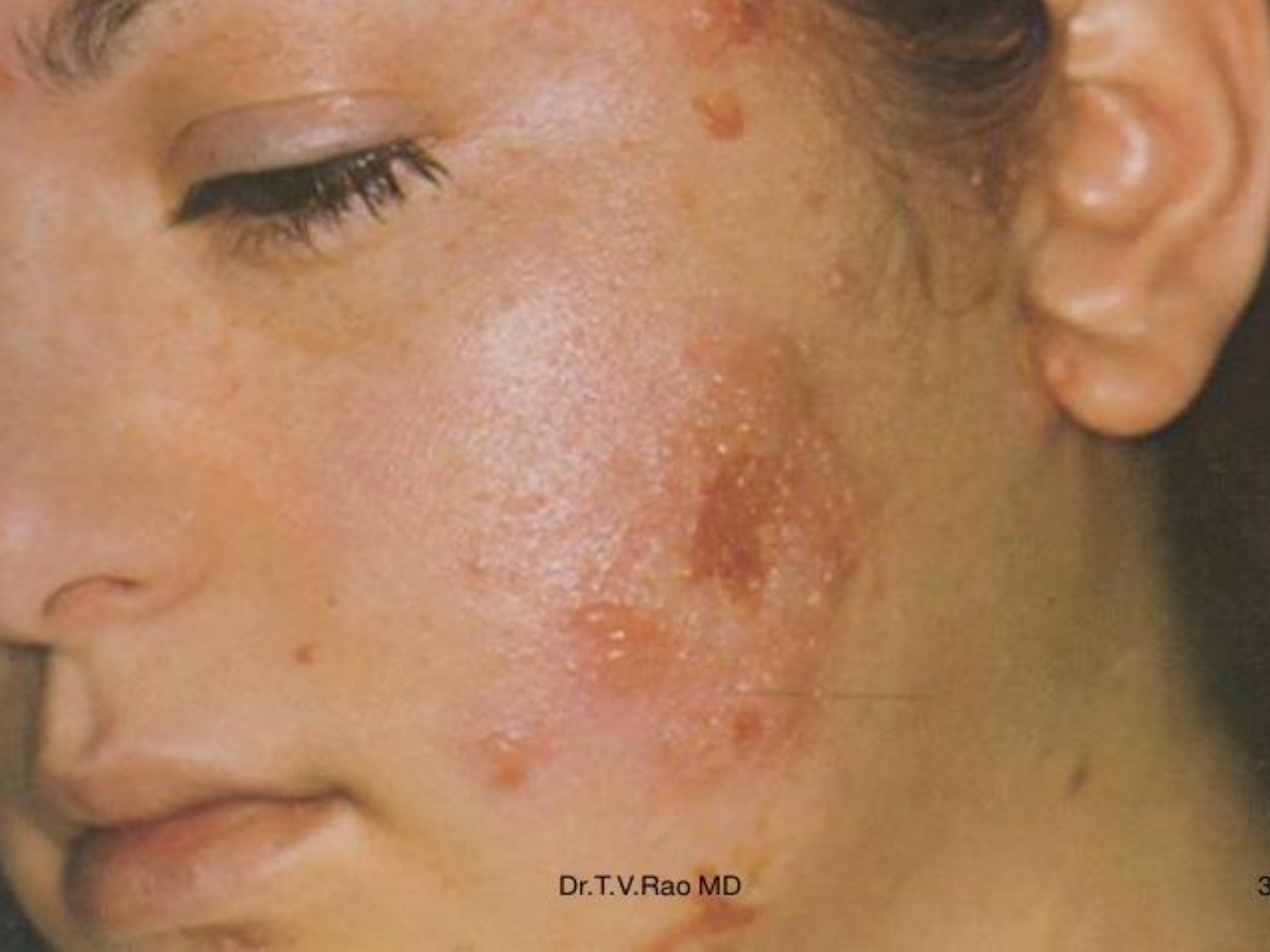
REACTIVATION

COLD
FEVER
SURGERY
UNKNOWN



Predisposition of Latent Infection in

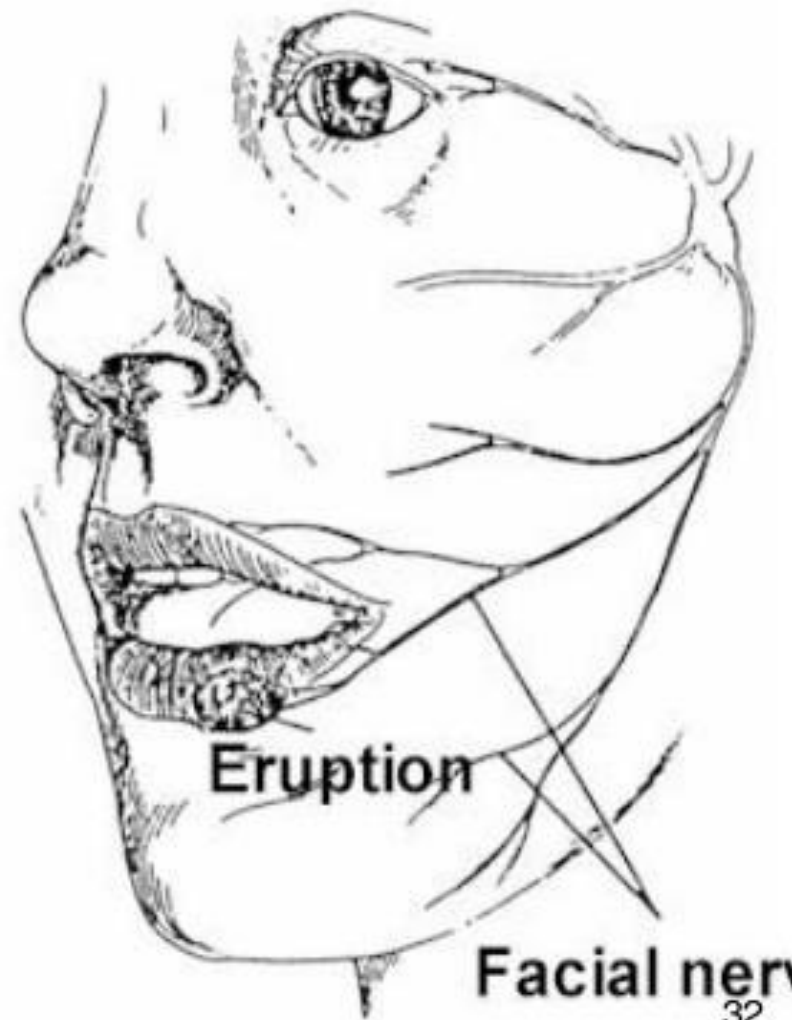
- Ganglion
 - Trigeminal HSV 1
 - Sacral HSV 2
 - Immunity.
 - Cell Mediated (CMI)
 - Predisposing Factors
 - Axonal Injury
 - Physical and Emotional stress
 - U V light
- 80% Adults harbour Antibodies to HSV



Recurrent Blisters in Herpes simplex 1



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Herpes lesions in the oral cavity



Eye Infections and Genital Infections.

- Corneal ulcerations
 pacifications
- Blindness
- Vesiculo ulcerative Lesions
 penis, Cervix, Vulva and
 Vagina.

Manifest with Painful lesions.

Herpes simplex 1 infecting eye



Skin Infections

- Infect abrasions
- Dentists, (Herpetic Whitlow) Health care workers,
- Eczema , Burns

Neonatal Herpes.

- In Uterus
- At Birth
- After Birth.
- Delivery By Caesarean Section

Reduces the Infection

Laboratory Diagnosis

- Microscopy,
- Antigen Detection
- DNA detection PCR.
- Viral Isolation.
- Serology

Laboratory Diagnosis

- Specimen: Vesicular fluid- Corneal scrapping

1- Direct Virus Demonstration:

a) L/M:

1. **Tzanck smear** - from the base of vesicles, 1% aq. soln. of toluidine blue 'O'
shows **multinucleated giant cells with faceted nuclei** & homogenously stained 'ground glass' chromatin (Tzanck cells)
2. **Giemsa stained smear** - intranuclear Cowdry type A inclusion bodies

- **2- Viral Isolation:** tissue culture: human diploid fibroblasts, human amnion, human embryonic kidney: CPC (syncytium formation) seen in 24-48 hrs.



- **3) Serology:** useful in the diagnosis of primary infection, Ab (IgM) detection by ELISA, NT or CFT.

PICORNAVIRUSES

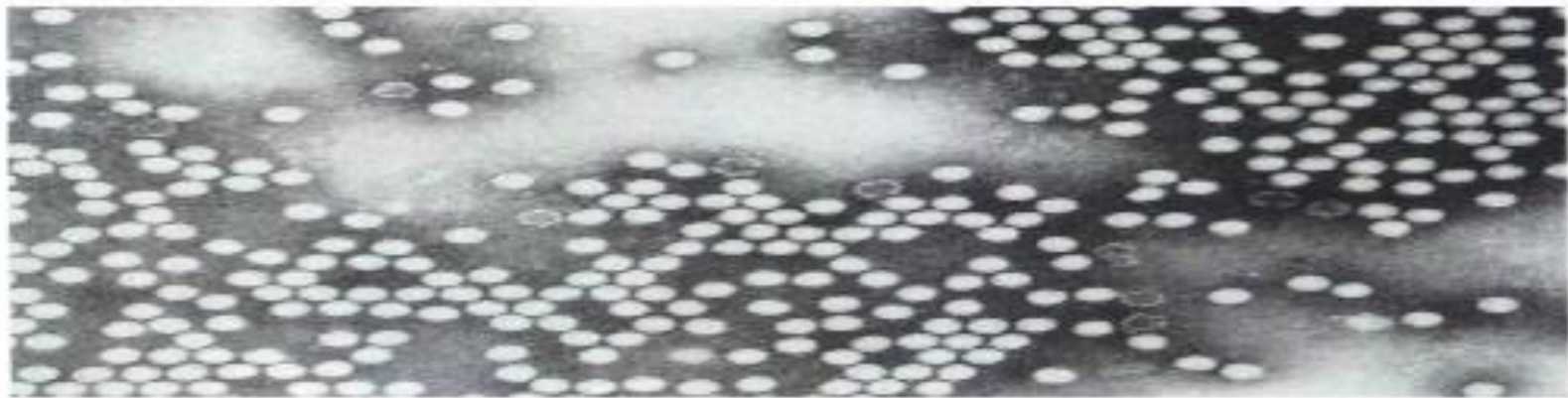


FIGURE 57-1. Electron micrograph of poliovirus. (Courtesy Centers for Disease Control and Prevention, Atlanta.)



PROPERTIES

Structure and composition

30 nm, icosahedral

plus-strand RNA, 7.2-8.4 kb

RNA is poly adenylated

VP1, VP2, VP3, VP4 **structural** proteins

VP4 interacts with viral RNA

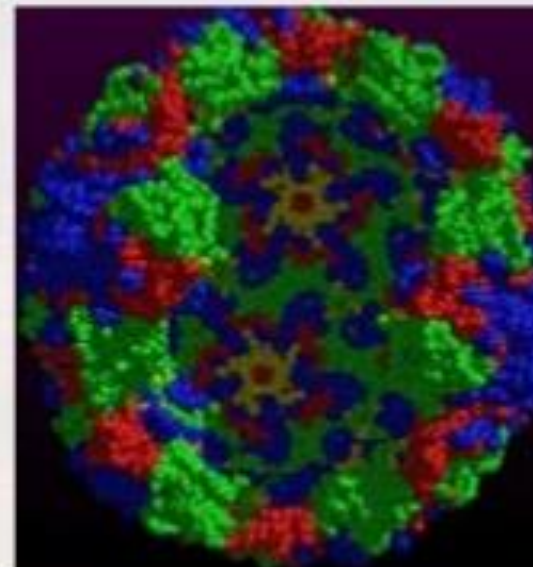
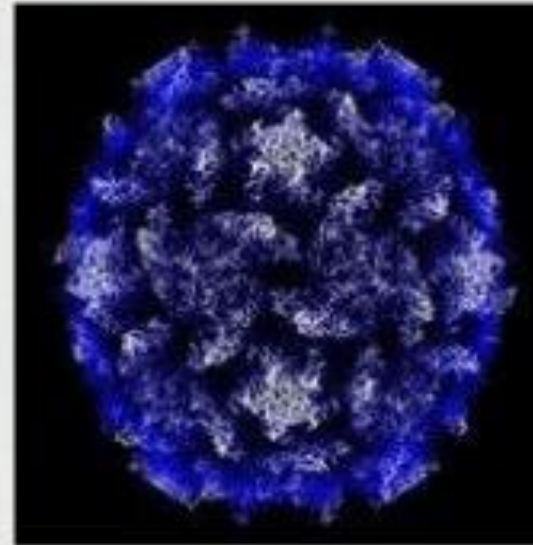
2A, 2B, 2C proteases

3A, 3B, 3C, 3D RNA replication

Nonenveloped

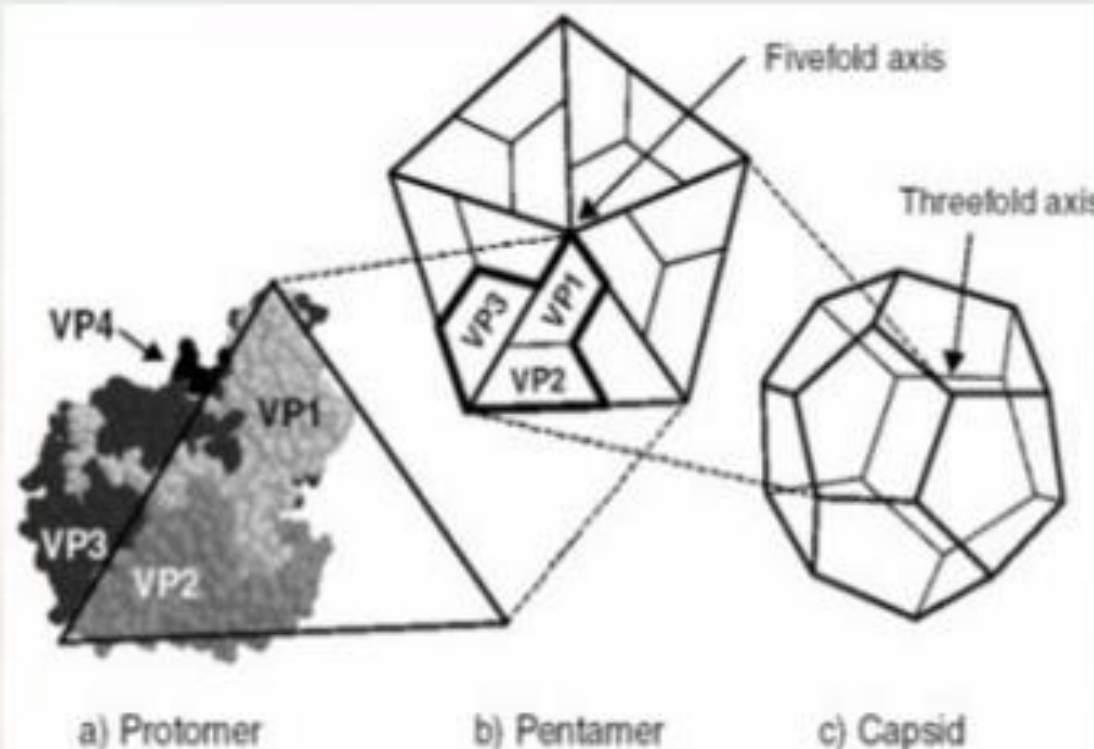
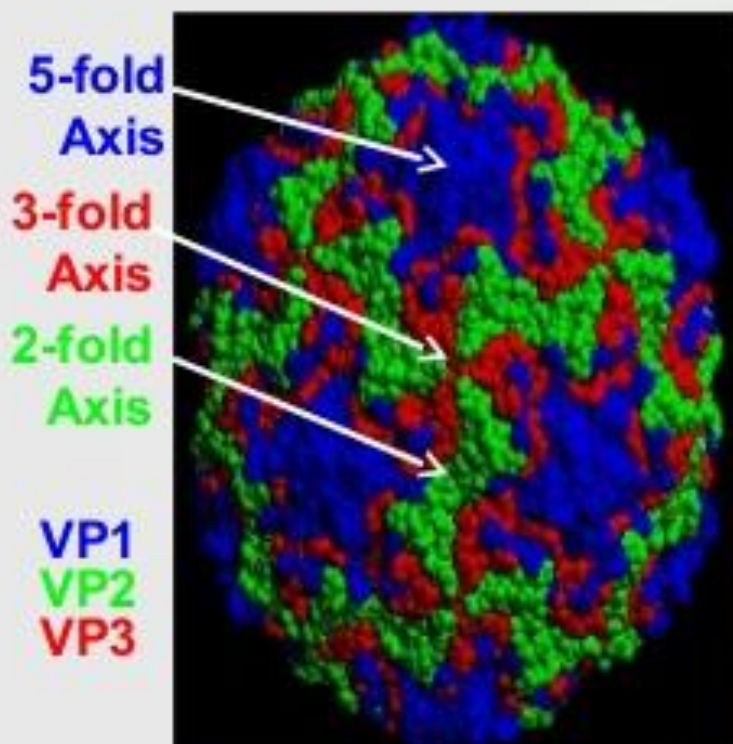
Cytoplasmic replication

Resistant to pH 3 to 9 (except for
Rhinoviruses).

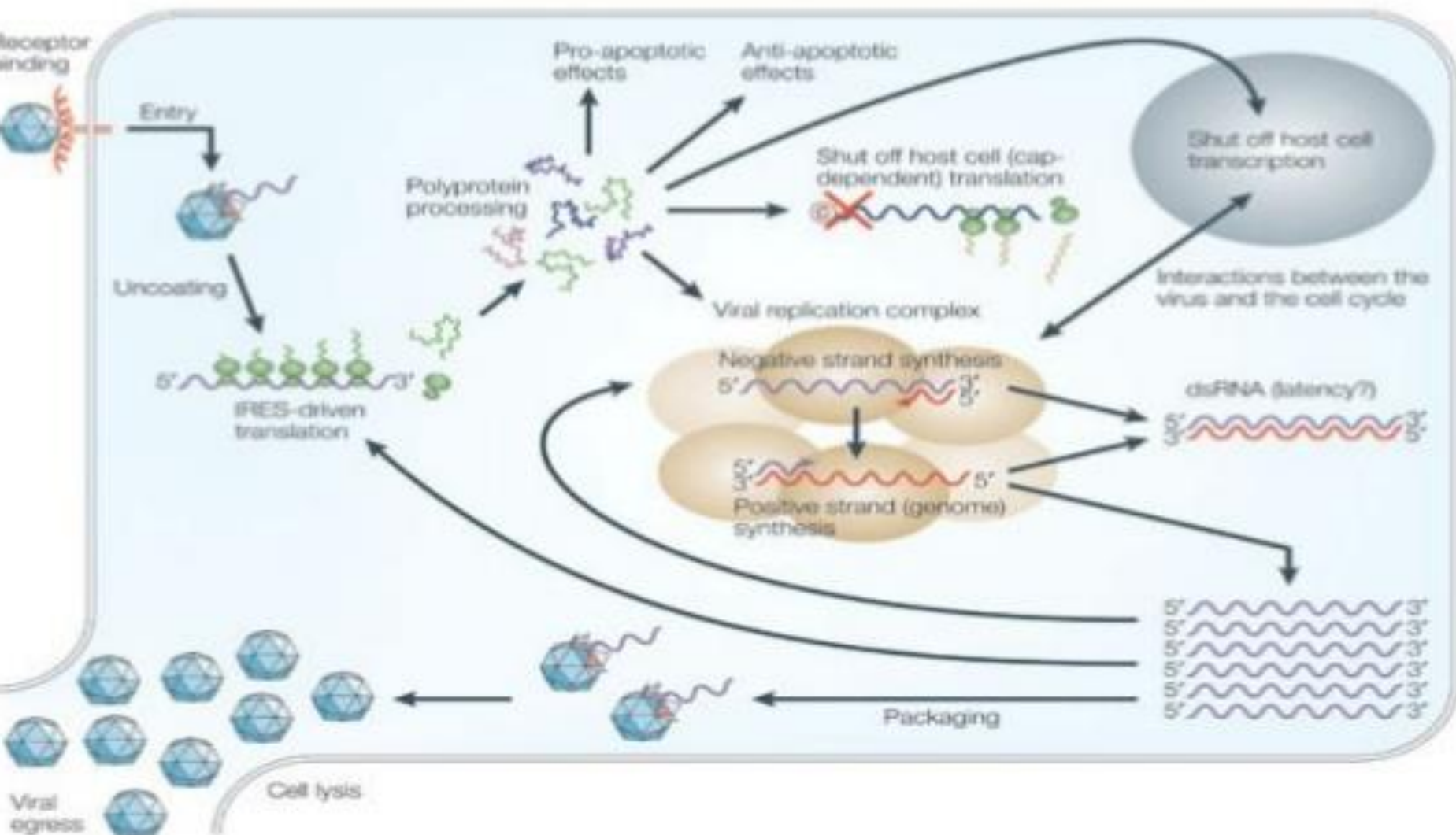


PICORNAVIRUS STRUCTURE

- The Basic capsid building block is a protomer that consists of one copy each of VP1, VP2, VP3 & VP4. VP1, VP2 & VP3 are on the virion **surface**, with VP4 being internal. VP1, VP2 & VP3 have no sequence homology, but have the same topology.

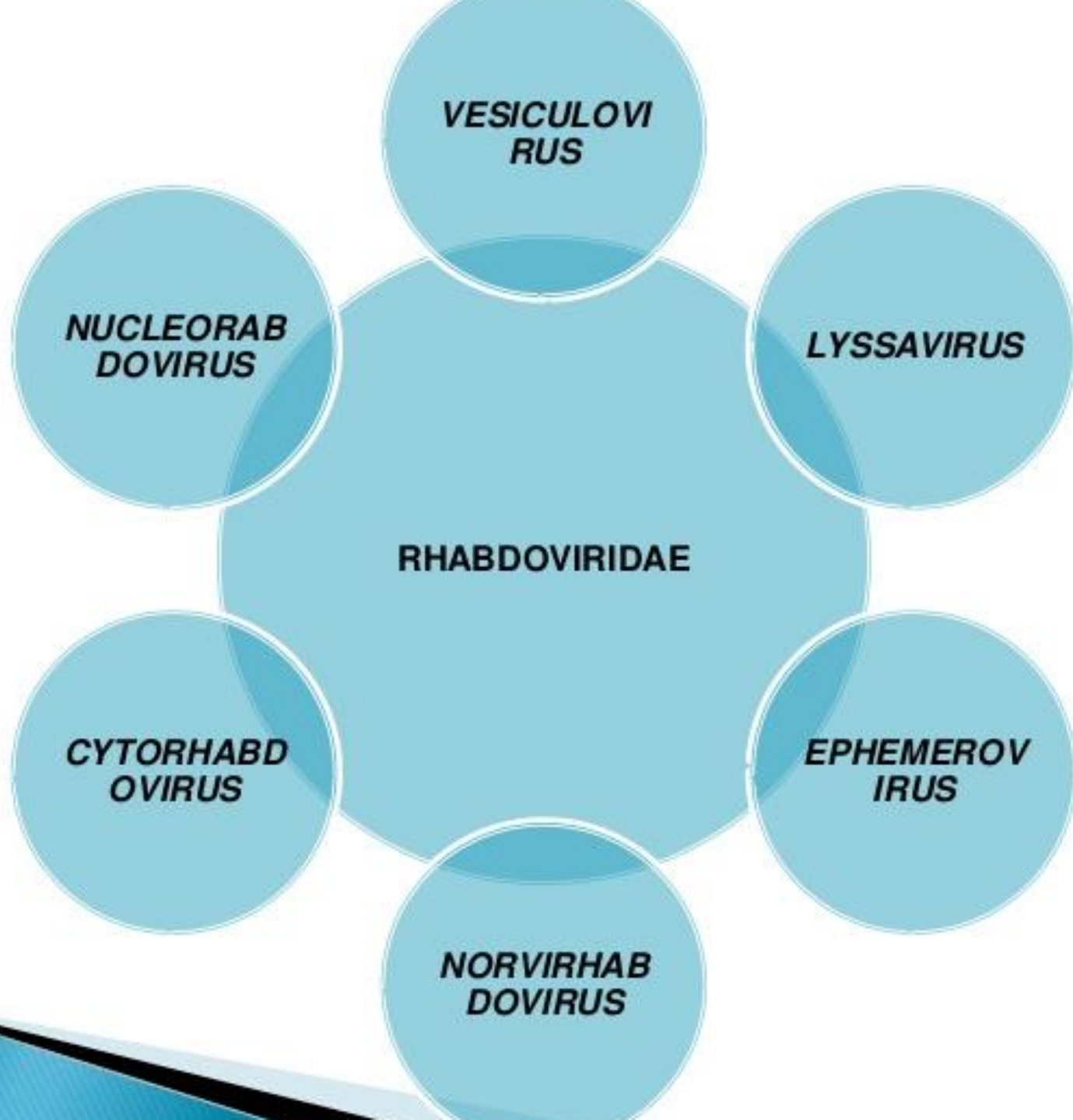


PICORNAVIRUS REPLICATION CYCLE



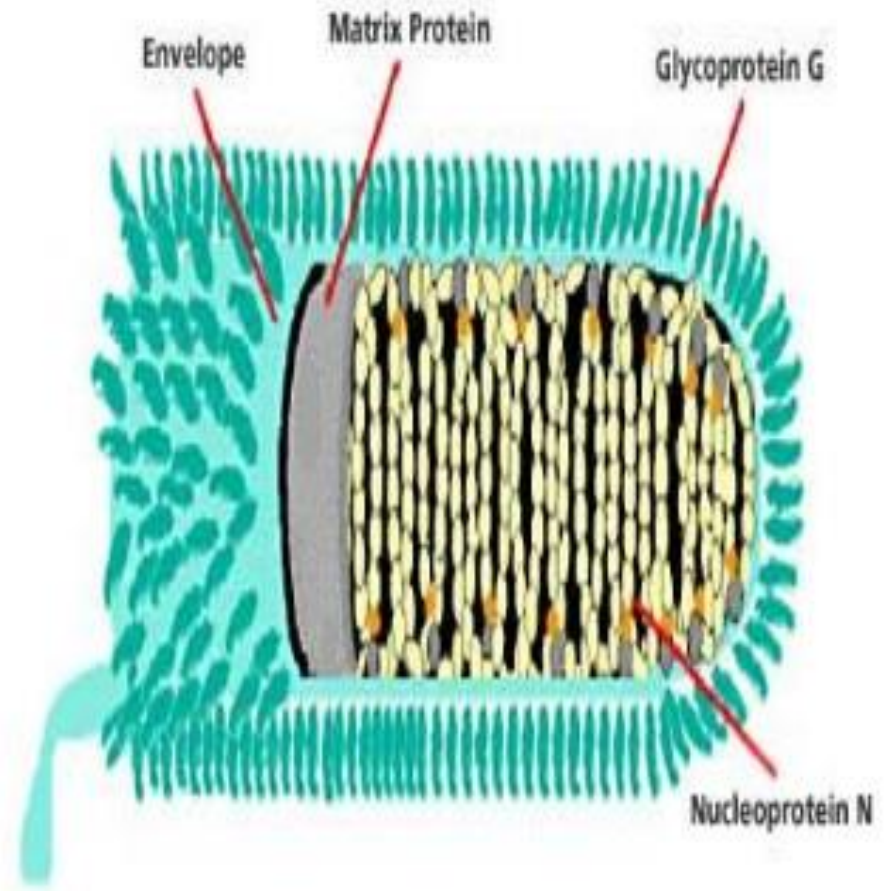
RHABDOVIRUSES:

- ▶ ***Mononegavirales* order**
- ▶ ***are negative, single stranded, monopartite RNA viruses***
- ▶ **capsid is roughly bullet shaped**
- ▶ **genome, about 11-15 kb in size**
- ▶ **Encodes for 5 to six proteins.**
- ▶ **SIX genera**



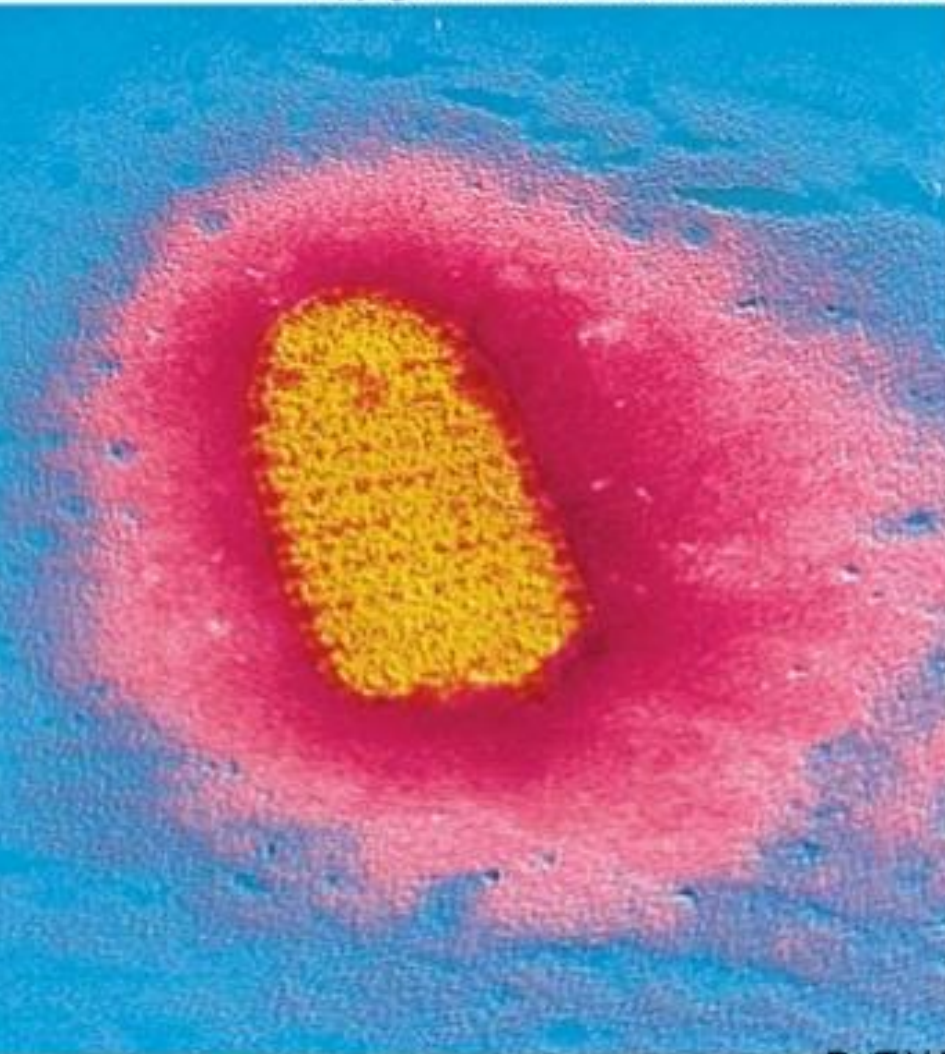
Rabies virus

- Bullet shaped virus
- Size is 180 x 75 nm
- Has Lipoprotein envelop
- Knob like spikes /Glycoprotein S
- Genome un segmented
- Linear negative sense RNA

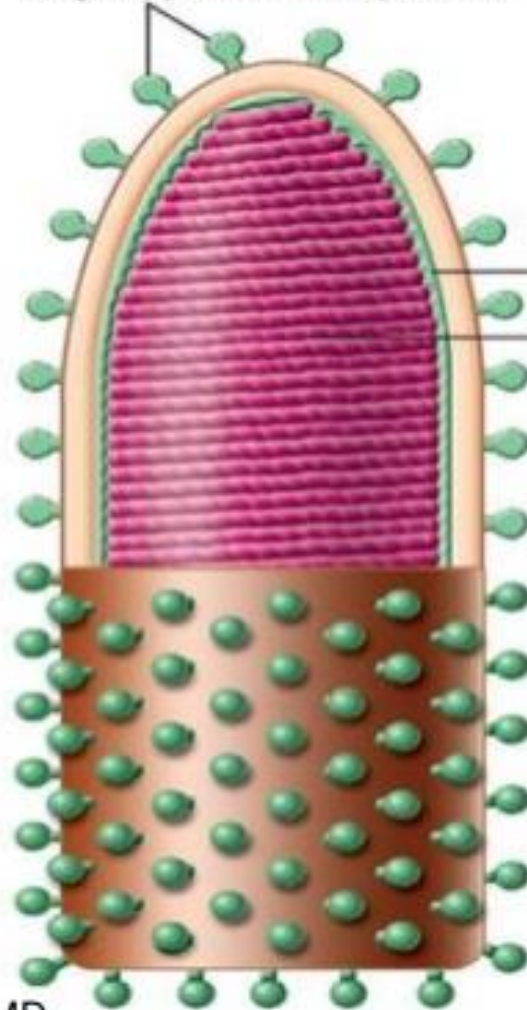


Structure of the rabies virus

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Glycoprotein spikes



Matrix protein

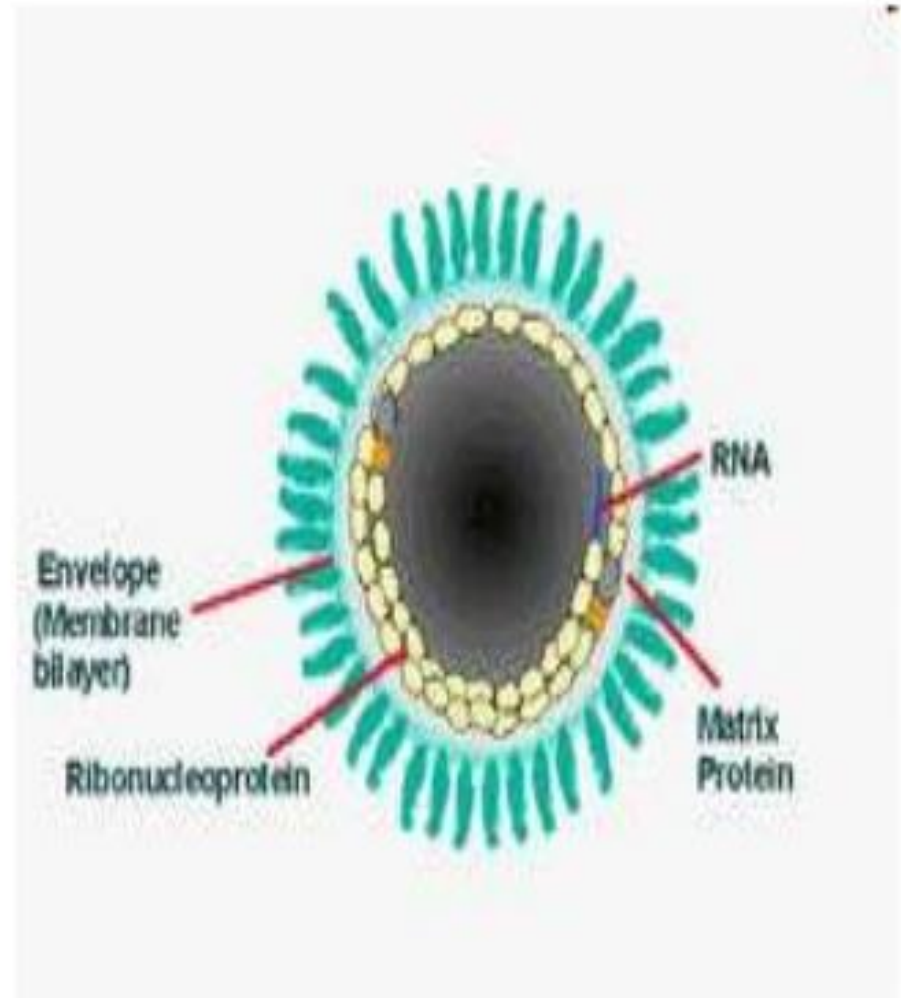
Nucleocapsid

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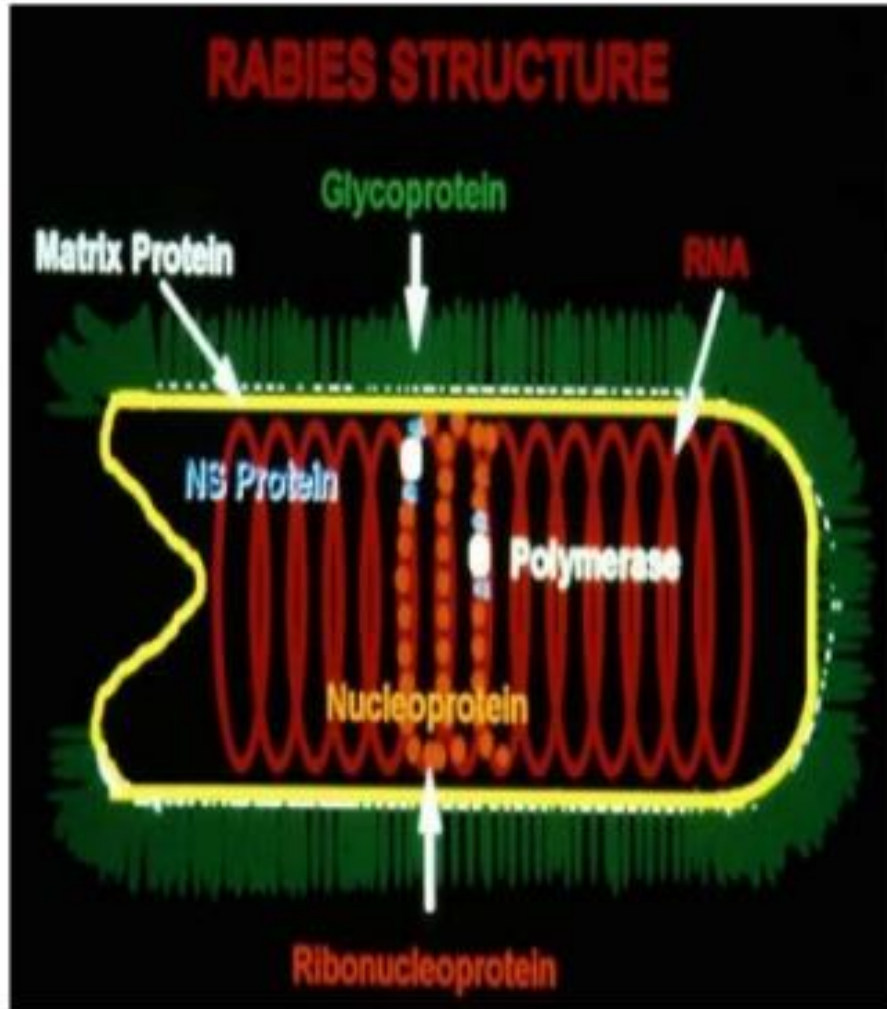
(b)

Fixed Virus

- One whose virulence and incubation period have been stabilized by serial passage and remained fixed during further transmission.
- Rabies virus that has undergone serial passage through rabbits, thus stabilizing its virulence and incubation period



Street Virus

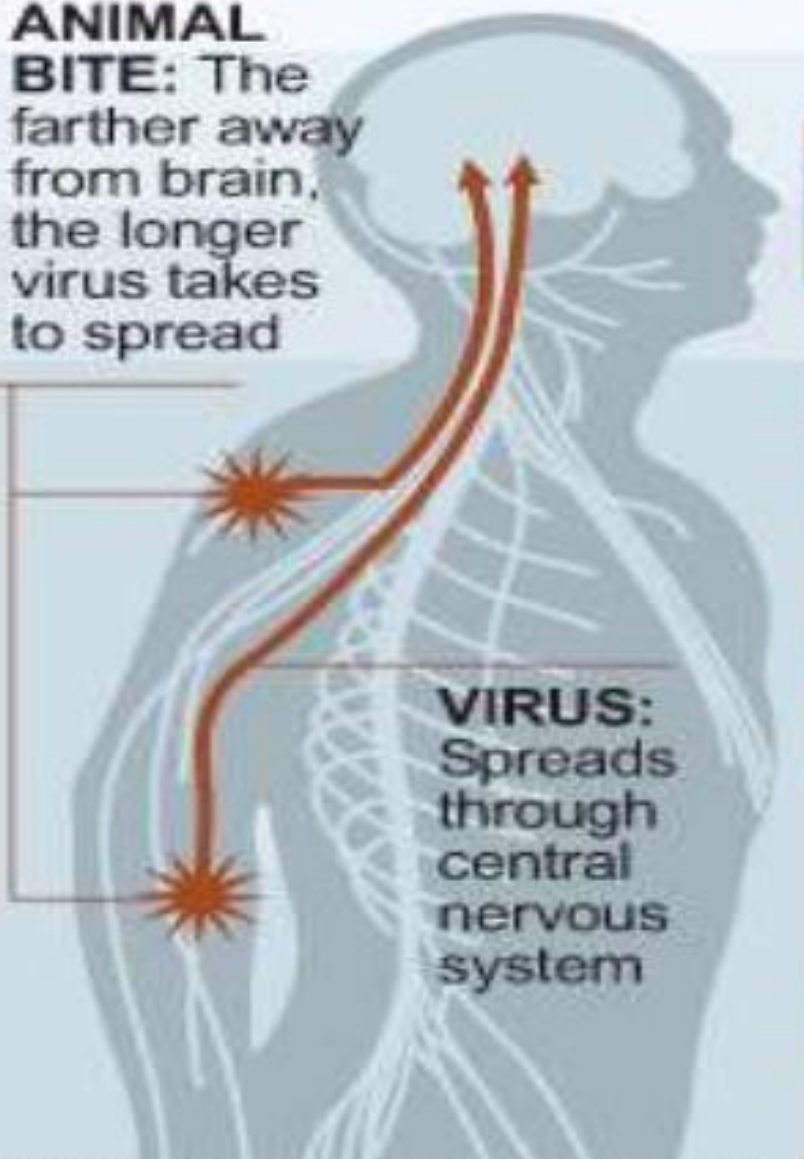


- Virus from a naturally infected animal, as opposed to a laboratory-adapted strain of the virus
- The virulent rabies virus from a rabid domestic animal that has contracted the disease from a bite or scratch of another animal.

Rabies

How it spreads

ANIMAL BITE: The farther away from brain, the longer virus takes to spread



VIRUS:
Spreads
through
central
nervous
system

Common carriers of rabies

Infected animals: Show no fear for humans; act very agitated



Dog: Another common rabies source

Symptoms in humans

- Fever, depression
- Agitation
- Painful spasms followed by excessive saliva
- Death within a week without vaccine



Treatment:
Hospitalization,
immune globulin
injections, anti-
rabies vaccine



Foaming at mouth after drinking:
Produced by
spasms in throat

Pathogenesis of Rabies

- Bite by Rabid dog or other animals
- Virus are carried in saliva virus deposited on the wound site.
- If untreated 50% will Develop rabies.
- Rabies can be produced by licks and corneal transplantation.
- Virus multiply in the muscle ,connective tissue, nerves after 48 – 72 hours.
- Penetrated nerve endings.

PATHOGENESIS

Live virus → Epidermis, Mucus membrane

Peripheral nerve

centripetally

CNS (gray matter)

centrifugally

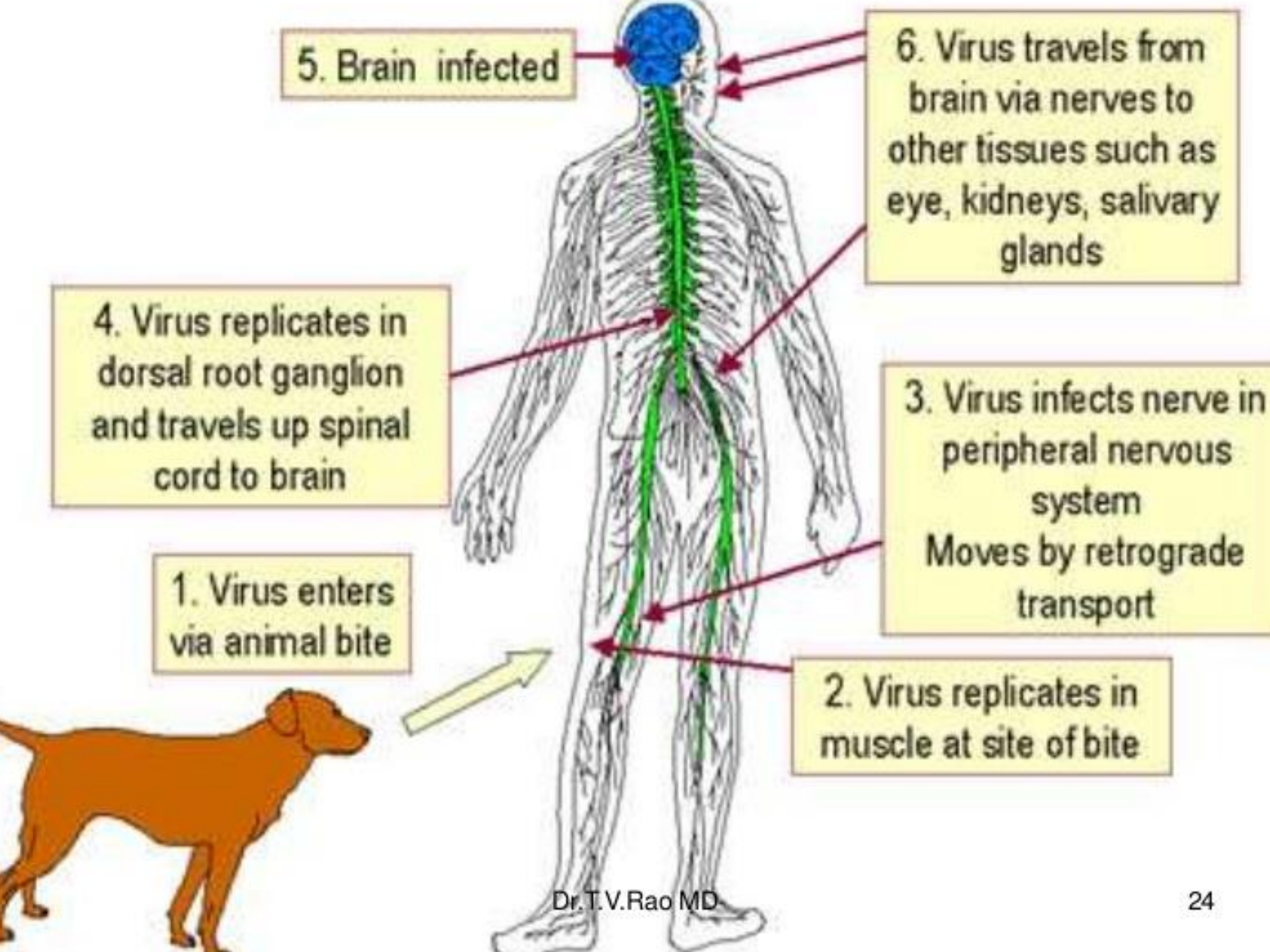
Other tissue (salivary glands,...)

Brain inflammation



Virus transmitted by
infected saliva
through bite
or wound



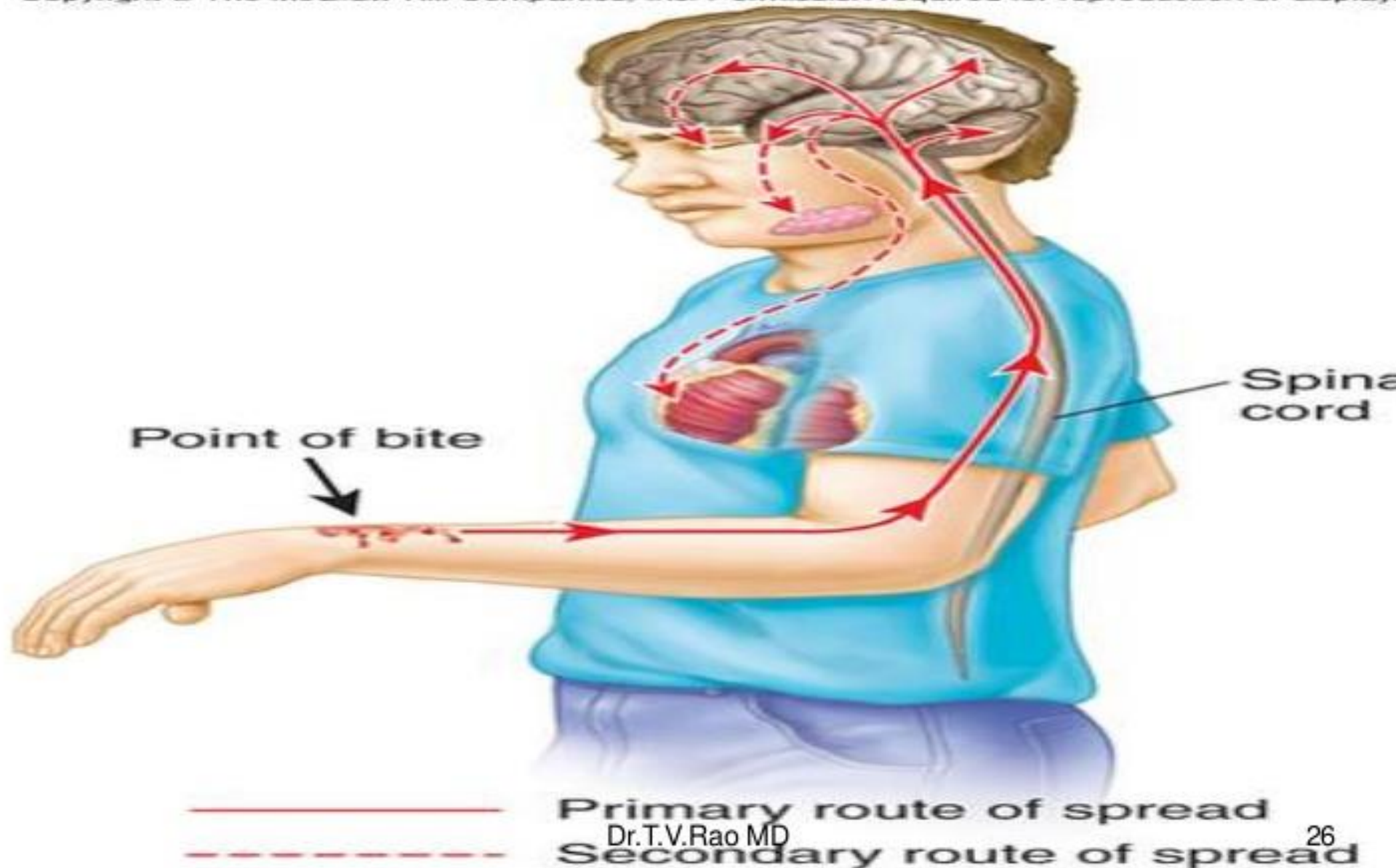


Spread of Virus

- From Brain virus spread to
Salivary glands,
Conjunctival cell released
into tears
Kidney
Lactating glands and Milk
after pregnancy

Pathologic pictures of Rabies

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Pathogenesis

- Incubation 1 – 3 months.
- **May be average from 7 days to 3 years.**
- Stages of the disease.

Prodrome

Acute encephalitis.

Coma / Death.

Category as per WHO

- **Category I:** touching or feeding suspect animals, but skin is intact
- **Category II:** minor scratches without bleeding from contact, or licks on broken skin
- **Category III:** one or more bites, scratches, licks on broken skin, or other contact that breaks the skin; or exposure to bats

Symptoms

- Headache, fever, sore throat
- Nervousness, confusion
- Pain or tingling at the site of the bite
- **Hallucinations**
 - Seeing things that are not really there
- **Hydrophobia**
 - “Fear of water” due to spasms in the throat
- **Paralysis**
 - Unable to move parts of the body
- Coma and death

CLINICAL MANIFESTATIONS

1 – Non specific prodrome

2 – Acute neurologic encephalitis

Acute encephalitis

Profound dysfunction of brainstem

3 – Coma

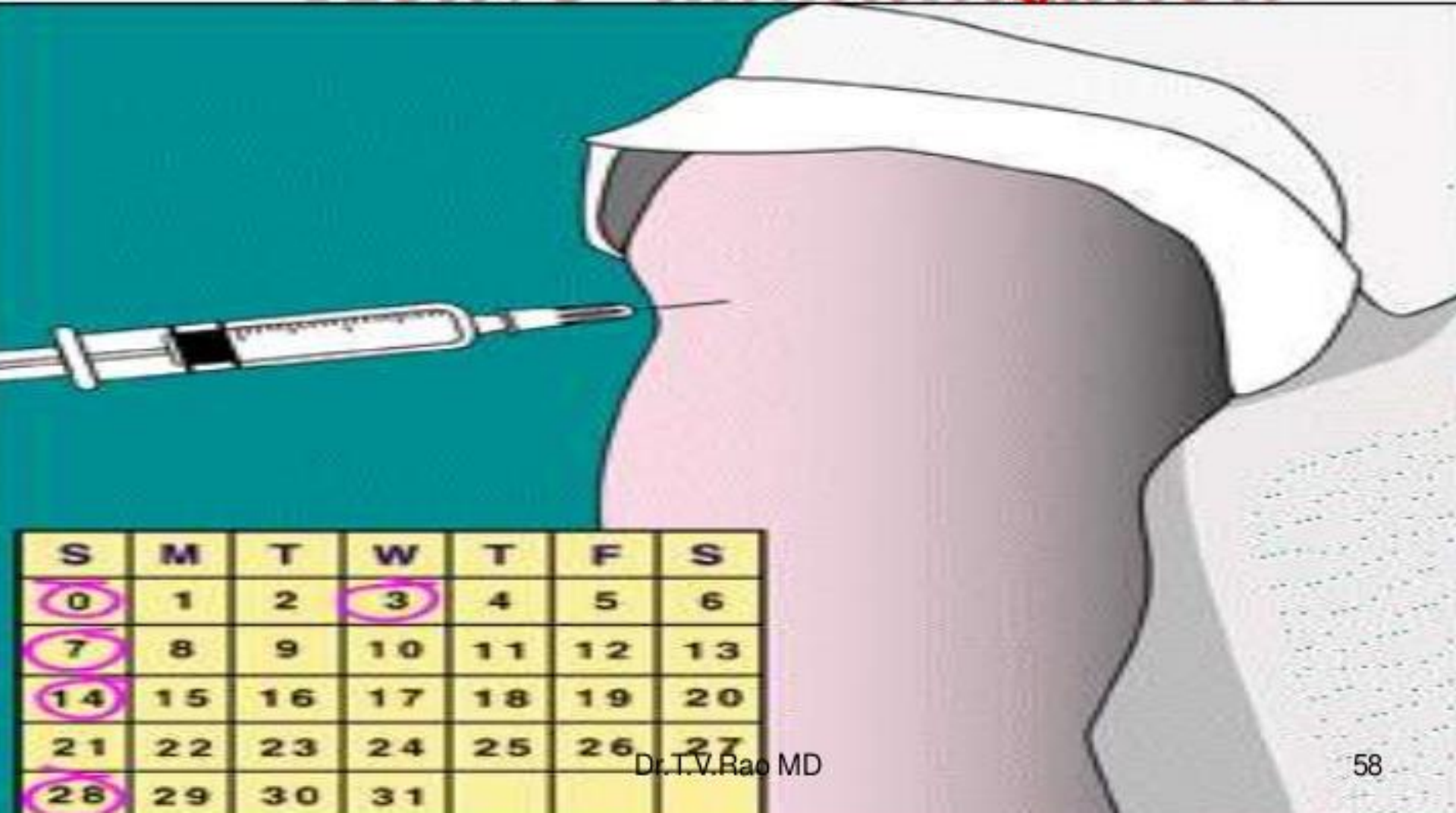
4 - Death (Dr.T.V.Rao MD Rare cases → recovery)³³

Post exposure Prophylaxis

- The vaccination is given on
0, 3, 7, 14, 30, and 90th day
Immunity lasts for 5 years
Injected on deltoid region IM/SC
Not to be given in the gluteal
region

POSTEXPOSURE PROPHYLAXIS

– *Active immunization*



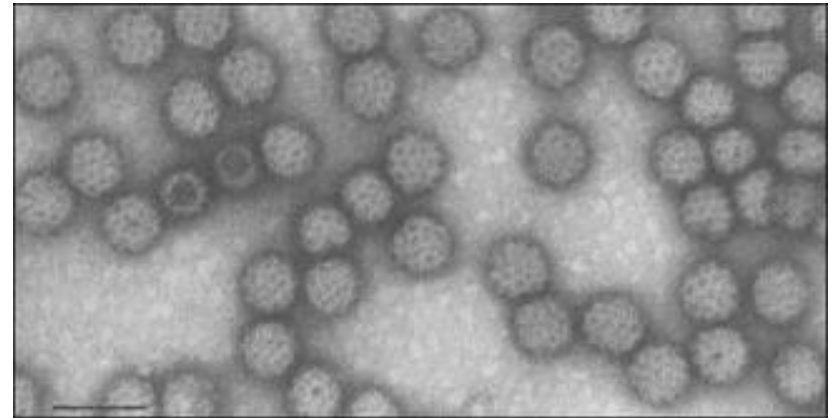
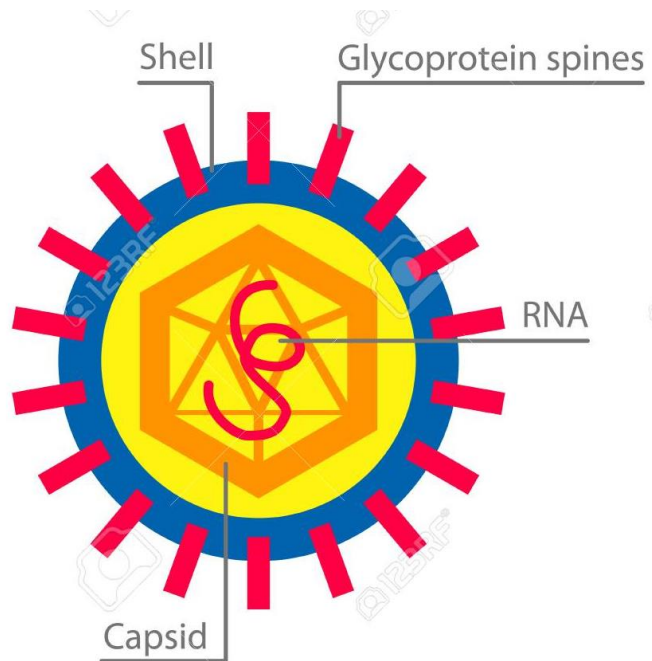
Arboviruses and Roboviruses

- **Arboviruses (arthropod born viruses)** are infected with blood-sucking arthropods (mainly mosquitoes and ticks).
- The main hosts for **roboviruses (rodent born viruses)** are rodents, the infection occurs in a non-transmissible way - through the biological excrement of rodents.

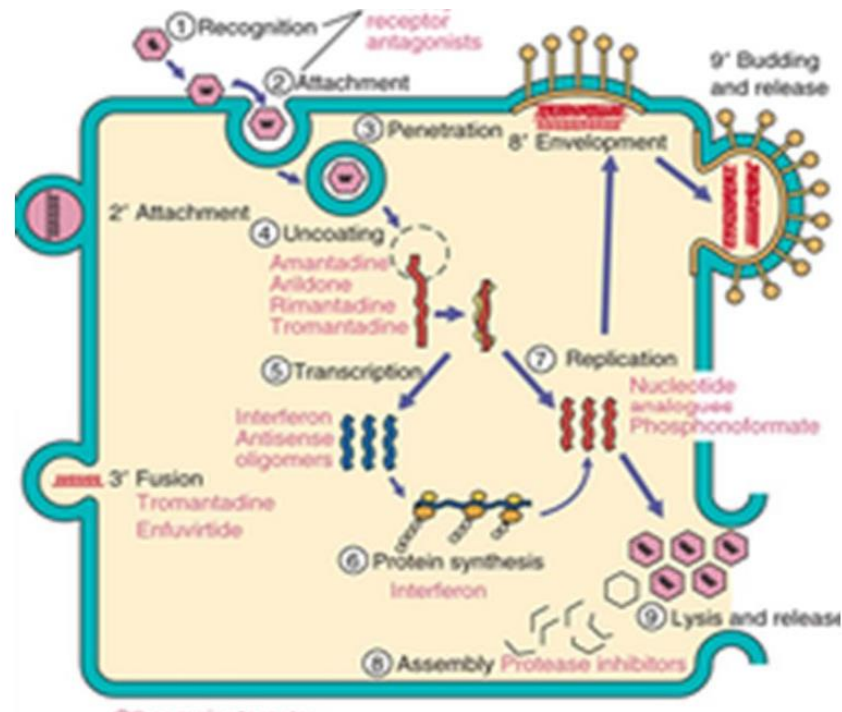
Arboviruses and Roboviruses

- Arboviruses include:
- Toqaviridae,
- Flaviviridae,
- Arenaviridae,
- Bunyaviridae
- Roboviruses include:
- *Bunyaviridae* (genus *Hantavirus*),
- *Arenoviridae*
- *Filoviridae*

Togaviridae



Reproduction of togaviruses

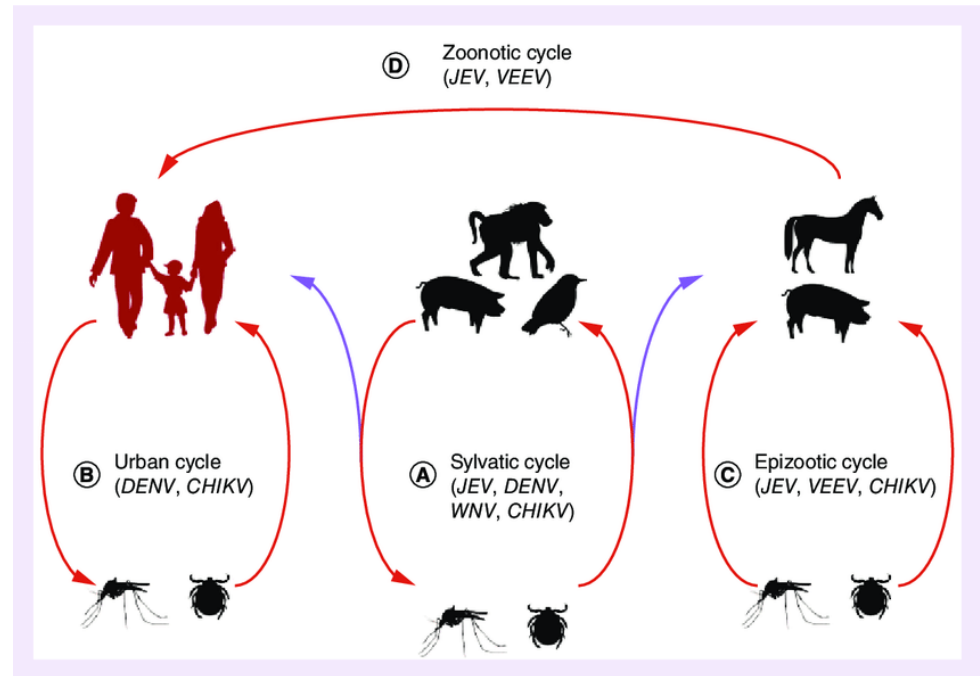


Classification of togaviruses

- Two genus of Togaviridae – **Alphavirus** and **Rubivirus** have clinical importance in human pathology
- ***Alfa viruses*** belong to arboviruses and caused disease that transmitted by arthropods
- ***Rubivirus*** not belong to arboviruses and transmitted by ear dropled mechanism

Sources of Infection and transmission ways

- Alphaviruses cause naturally occurring zoonotic diseases.
- In natural habitats, the reservoir of the virus are vertebrates - birds, rodents, primates and other animals.
- In natural habitats, people become infected through the bite of arthropods.
- The virus multiplies in the tissues and organs of arthropods, including the salivary glands.



Pathogenesis of Alfavirus infections

- Viruses that enter the body through the blood-sucking of arthropods first multiply in the local subcutaneous tissue and regional lymph nodes.
- The viruses then pass into the bloodstream and spread throughout the body, and depending on the nature of the causative agent, their subsequent proliferation occurs in monocytes and macrophages, vascular endothelium, lungs, liver, muscles, etc. possible.
- Neurotropic viruses enter the central nervous system and cause degenerative changes in brain cells, resulting in encephalitis.

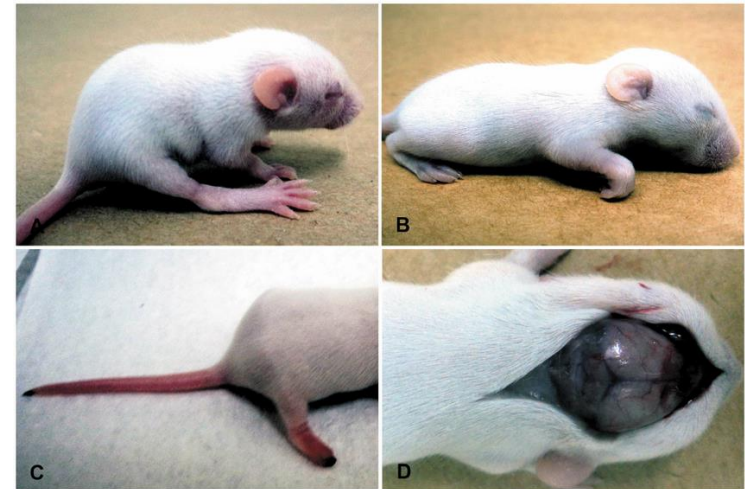
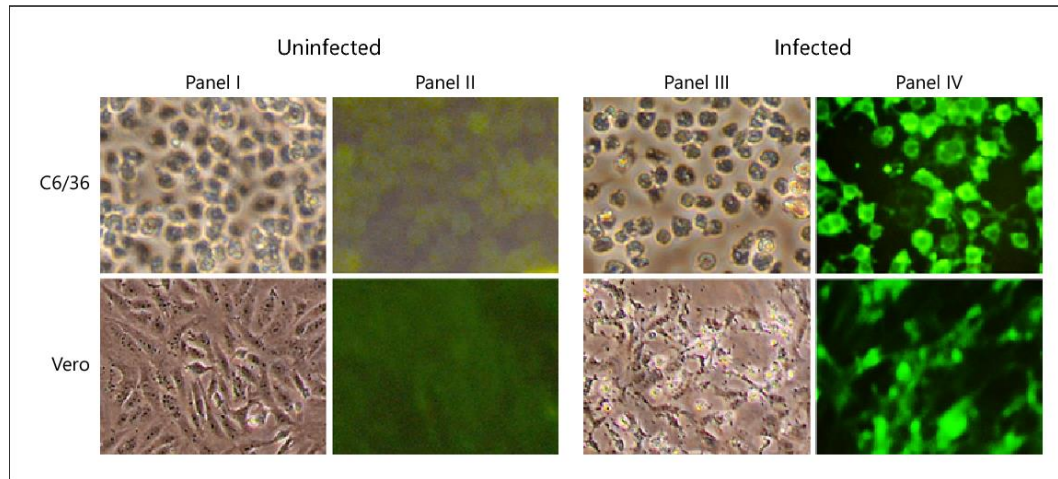
Clinical forms of alphavirus infections

- ***Sindbis fever***. The causative agent of the virus was obtained from mosquitoes in the village of Sindbis in Cairo (Egypt). The disease begins with fever, headache, arthralgia, skin rash and lasts 5-8 days. Although it has a benign end, it can become chronic and disability with the development of osteoarthritis.
- ***Semliki forest fever***. The causative agent of the virus was obtained from mosquitoes in the Semliki forest of Uganda. The disease is sporadic in humans and is manifested by fever, in some cases encephalitis and aseptic meningitis.
- ***Chikungunya fever*** is common in tropical and subtropical climates and is characterized by double-wave fever, intoxication, myalgia, severe joint pain, lymphadenopathy, maculopapular rash, and sometimes meningeal and hemorrhagic symptoms.
- ***Equine encephalomyelitis***. Diseases in humans are mainly found in many countries of the American continent (Brazil, Argentina, Mexico, USA, Canada, etc.), mainly accompanied by symptoms of encephalitis - darkening of consciousness, headache, fever, paralysis.

Microbiological diagnosis of alphavirus infections

- Viruses can be found in the blood in the early stages of the disease, and later in the cerebrospinal fluid. For this purpose, infantile white mice are infected intracerebral.
- Viruses can also be obtained by infecting appropriate cell cultures with pathological materials. Alphaviruses are identified in mice and cell cultures by NR, IFR, and ELISA.
- PCR is used in the diagnosis of some diseases.
- In the serum of patients it is possible to identify antibodies to the virus neutralizing and antihemagglutinin, which appear a few days after the disease and persist for many years. The simplest way to determine these antibodies is the inhibition of hem agglutination test. Determination of virus-specific IgM in cerebrospinal fluid is considered a more sensitive test.
- The diagnosis is confirmed by the fact that the titer of antibodies in the blood serum taken at the beginning of the disease and 2-3 weeks later increased by 4 times or more.

Microbiological diagnosis of alphavirus infections



Flaviviridae

- >68 viruses
- small
- spherical
- enveloped
- SS RNA
- cross-related

Flaviviruses

- Yellow Fever virus
- Dengue viruses
- St. Louis encephalitis virus
- Japanese encephalitis virus
- West Nile virus
- Murray Valley encephalitis virus, tick-borne encephalitis viruses and others

Yellow Fever

- vector: *Aedes aegypti*
- Latin America, Caribbean, Africa
- inapparent to severe infection (jaundice, hemorrhage, albuminuria)
- hepatic necrosis, Councilman and Torres bodies
- Dx: cell culture, serology, PCR, immunohistochemistry
- supportive treatment
- live attenuated 17D vaccine

Dengue Virus

- Causes dengue and dengue hemorrhagic fever
- Transmitted by mosquitoes
- Has 4 serotypes (DEN-1, 2, 3, 4)

World Distribution of Dengue - 2000



Aedes aegypti

- Dengue transmitted by infected female mosquito
- Primarily a daytime feeder
- Lives around human habitation
- Lays eggs and produces larvae preferentially in artificial containers with clean stagnant water



Clinical Characteristics of Dengue Fever

- Fever
- Headache
- Muscle and joint pain
- Nausea/vomiting
- Rash
- Hemorrhagic manifestations

Hemorrhagic Manifestations of Dengue

- Skin hemorrhages: petechiae, purpura, ecchymoses
- Gum bleeding
- Nose bleeding
- Gastro-intestinal bleeding: hematemesis, melena, hematochezia
- Hematuria
- Increased menstrual flow



Laboratory Methods for Dengue Diagnosis

- Virus isolation to determine serotype of the infecting virus
- IgM ELISA test for serologic diagnosis

Japanese encephalitis virus

- single serotype, 5 genotypes based on E protein
- Asia, including SEA
- cycle: birds - Culex mosquitoes - swine
- humans, horses

Japanese encephalitis

- 99% subclinical
- lethargy, behavioral changes, motor abnormalities
- Dx: CSF analysis, EEG, IgM ELISA, NT, HI, CF, PCR
- Rx: supportive
- Prevention: inactivated vaccine (3 yr protection)

Arenaviruses

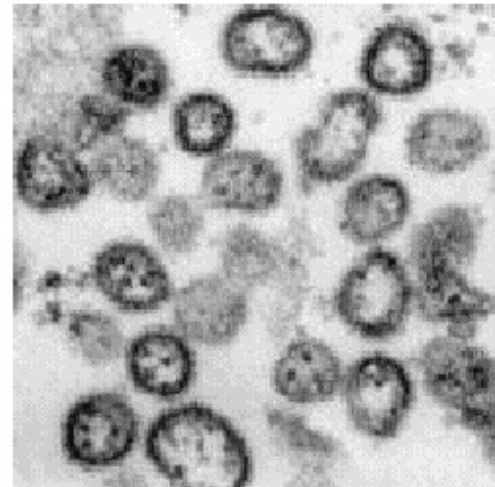




Introduction

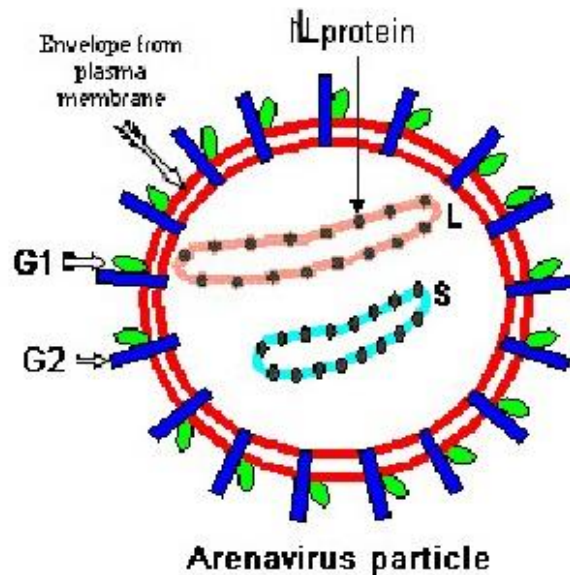


- Family = Arenaviridae
- Genus = Arenavirus
- Currently 22 recognized species
- 2 groups
 - Old World
 - New World
- Rodent – borne pathogens
- Important cause of VHF
- Host cell ribosomes are present in the viral particles are responsible for a “sandy” appearance on EM
- Hence the name Arenavirus (Latin: arena=sand)





Virology



- ssRNA virus “ambisense”
- Genome consists of 2 RNA segments (L) = Large (S) = Small
- (L) encodes RNA-polymerase & Zn-binding protein
- (S) encodes NP and GPC
- Virions are spherical to pleomorphic
- Enveloped
- Average diameter = 120nm
- Envelope covered with 8-10nm long projections



Classification



2 groups



New World

Tacaribe Serocomplex

17 species

3 clades (A,B,C)



Family: *Muridae*

Subfamily: *Sigmodontinae*

American rodents *



Old World

Lassa-LCM Serocomplex

5 Species



Family: *Muridae*

Subfamily: *Murinae*

Eurasian rodents



Reservoirs



Rodent - mastomys sp

- Usually one species, less often 2 closely related species
- Chronic mild infection
- Life long shedding of virus
- Except Tacaribe virus *



*Fruit-eating bat – Artibeus sp



Transmission

- Rodent – Rodent

- Vertical

- Horizontal (aerosolized urine, faeces, saliva, bites)

- Rodent – Human

- Aerosolized secretions

- inoculation via cuts, bites

- contaminated fomites, food

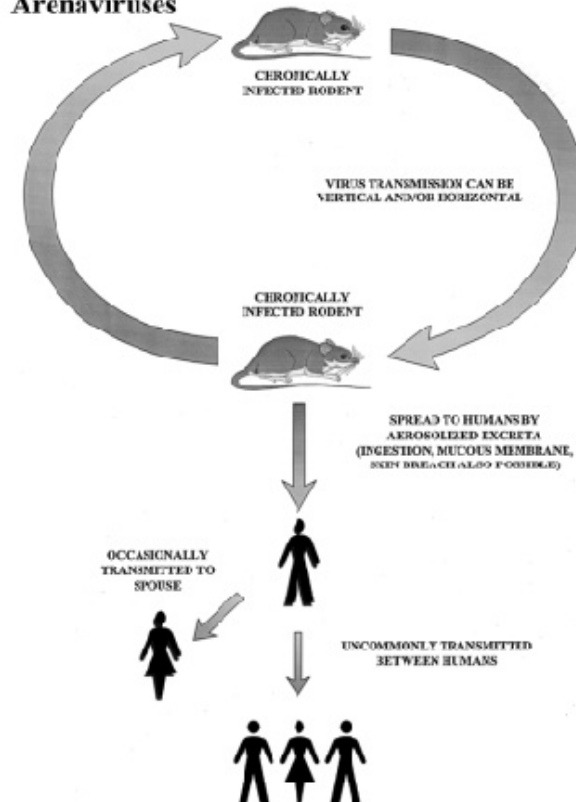
- Rodent consumption

- Human – Human

- contaminated secretions, sexual

- Inoculation

Arenaviruses





Clinical presentation



Table 2 Clinical stages of Lassa fever (adapted from McCarthy 2002¹¹)

Stage	Symptoms
1 (days 1-3)	General weakness and malaise. High fever, $>39^{\circ}\text{C}$, constant with peaks of $40\text{--}41^{\circ}\text{C}$
2 (days 4-7)	Sore throat (with white exudative patches) very common; headache; back, chest, side, or abdominal pain; conjunctivitis; nausea and vomiting; diarrhoea; productive cough; proteinuria; low blood pressure (systolic <100 mm Hg); anaemia
3 (after 7 days)	Facial oedema; convulsions; mucosal bleeding (mouth, nose, eyes); internal bleeding; confusion or disorientation
4 (after 14 days)	Coma and death





Laboratory Diagnosis



Test	Specimen	Comments
Culture	Blood, CSF, Tissue	Cell Culture: Vero, Vero B6, BHK Animal: suckling mice, hamsters, guinea pigs
EM	Blood, urine, tissue	
Antigen detection	Blood	ELISA/IMF
Immunohistochemistry	Tissue	Liver, spleen, skin, kidney
RT-PCR	Blood, tissue	
Serology (IgG/IgM)	Serum	ELISA

Bunyaviridae

Structure

- Spherical enveloped particles
 - 90-100 nm
- virus encoded transcriptase
- 2 external glycoproteins
- multipartite - 3 segments of SS RNA
 - antisense



Classification

- Genera

- *Bunyaviruses*
- *Phlebovirus*
- *Nairovirus*
- *Uukuvirus*
- *Hantavirus*

- Immunologically

- 35 serogroups
- 300 types & subtypes

Serious Bunya Virus Diseases

- Crimean Congo Hemorrhagic Fever Virus
- Rift Valley Fever Virus
- La Crosse Virus
- Hanta Virus

Pathogenesis

- Early
 - fever & viremia
- Late
 - encephalitis
 - retinitis
 - renal involvement



Epidemiology

- Determined by distribution of vector and mammalian host
- Humans: accidental dead end hosts

Diagnosis

Presumptive

- febrile illness
- geographic site of exposure
- Sean
- vectors

Confirmation

- virus isolation
- Virus specific IgM
- Rise in antibody titer

Control

- Control Vectors

- arthropods
- rodents

- Vaccination

- Humans: Crimean Congo
- Sheep & Cattle : Rift Valley

Crimean Congo Hemorrhagic Fever Virus

- Headache
- pain in limbs
- bleeding from multiple orifices

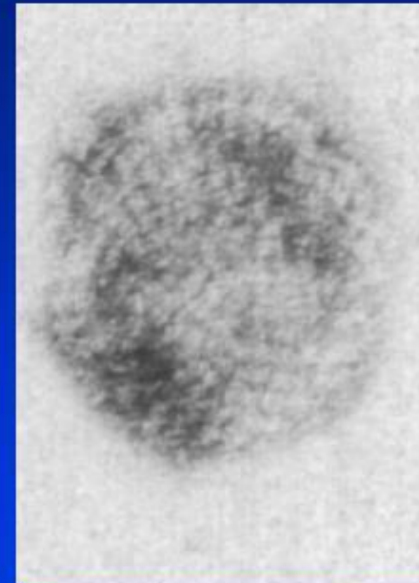
Hanta Virus

- Rodents - transmission to humans via aerosolized excretions and bites. A wide variety of biting insects.



Hanta Virus

- Lipid envelope
 - susceptible to most disinfectants.
 - Need to lower pH < 5



Hanta Virus

- tripartite negative-sense RNA
 - L, >> viral transcriptase
 - M >> envelope glycoproteins
 - S, nucleocapsid protein



Hantavirus Pulmonary Syndrome

- febrile prodrome
- followed by
 - non-cardiogenic pulmonary edema,
 - hypotension
 - shock