AZERBAIJAN MEDICAL UNIVERSITY
DEPARTMENT OF MEDICAL MICROBIOLOGY and IMMUNOLOGY

LESSON 17.

Microbiology diagnosis of infections, caused by arboviruses group (families of *Togaviridae, Flaviviridae, Arenaviridae, Bunyaviridae, Reoviridae* and *Filoviridae*) and *Rhabdoviridae* family

FACULTY: ***General Medicine***
SUBJECT: ***Medical microbiology - 2***

Discussed questions:

1. Togaviridae family, general properties (classification, morphology, reproduction, antigen structure, durability).
• Alphavirus genus, general properties, pathogenesis and clinical forms (Sindbis fever, Semlika forest fever, Chikungunya fever, horses encephalomyelitis). Microbiological diagnosis of alphavirus infections
2. Flaviviridae family, general properties (morphology, reproduction, durability, classification). Microbiological diagnosis, treatment and prevention of diseases caused by it (yellow fever, dengue fever, tick-borne encephalitis, Japanese encephalitis, West Nile fever, Omsk hemorrhagic fever)
3. Arenaviridae family, classification, general properties (morphology, reproduction, durability). Microbiological diagnosis, treatment and prevention of diseases caused by it (Lass fever, Lymphocytic choriomeningitis, South American hemorrhagic fevers)
4. Bunyaviridae family, general properties (morphology, reproduction, durability). Microbiological diagnosis, treatment and prevention of the diseases caused by it (California encephalitis, Mosquito fever (phlebotome fever), Rift Valley fever, Crimean-Congo hemorrhagic fever, Hantavirus pneumonia syndrome (HPS), hemorrhagic fever with renal syndrome (HHF)).
5. Reoviridae family, general properties (morphology, reproduction, persistence). Morpho-biological characteristics of rotaviruses, epidemiology, diseases caused by them, pathogenesis, microbiological diagnosis, treatment, prevention
6. Filoviridae family, classification, general properties (morphology, reproduction, antigen structure, persistence). Microbiological diagnosis of diseases caused by Marburg virus and Ebola virus
7. Rhabdoviridae family, general properties (classification, morphology, reproduction, antigen structure, persistence).
• General properties of the rabies virus (morphology, cultural and antigenic properties, types). Epidemiology, pathogenesis and clinical features of rabies. Microbiological diagnosis, specific prevention and treatment problems of rabies

Purpose of the lesson:

- To acquaint students with the morpho-biological characteristics of viruses belonging to the arbovirus group (Togaviridae, Flaviviridae, Arenaviridae, Bunyaviridae, Reoviridae and Floviridae families) and Rhabdoviridae family and to provide information on microbiological diagnostic methods of diseases caused by these viruses.

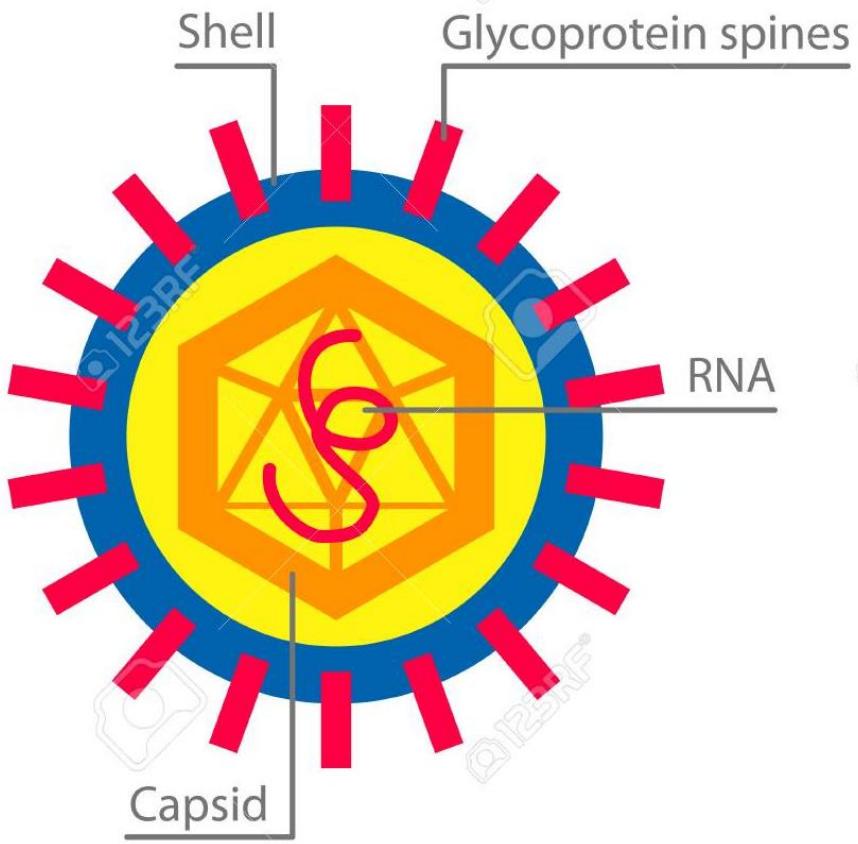
Arboviruses and Roboviruses

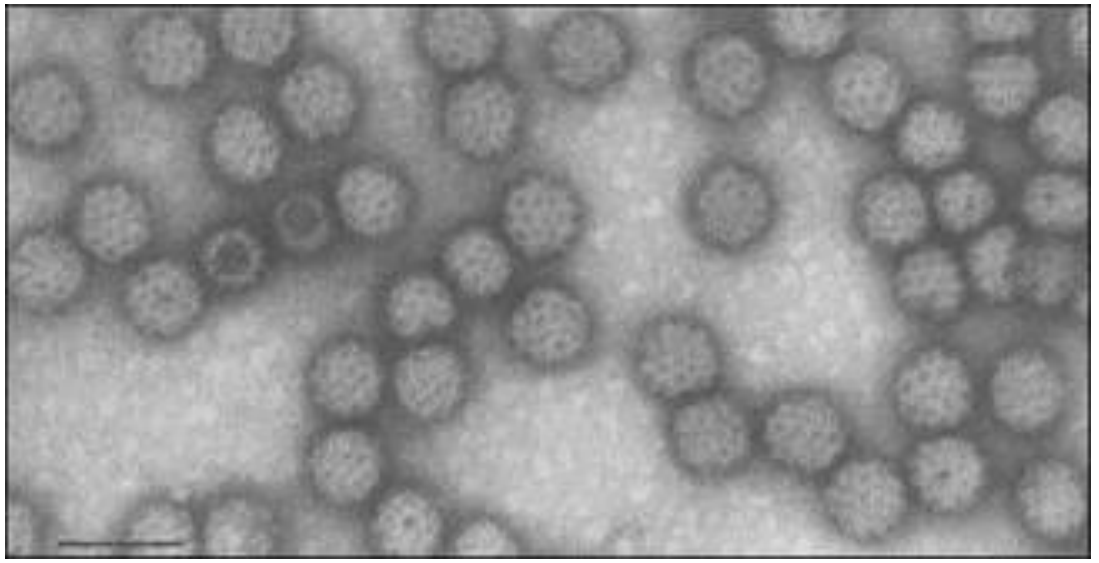
- **Arboviruses (arthropod born viruses)** are infected with blood-sucking arthropods (mainly mosquitoes and ticks).
- The main hosts for **roboviruses (rodent born viruses)** are rodents, the infection occurs in a non-transmissible way - through the biological excrement of rodents.

Arboviruses and Roboviruses

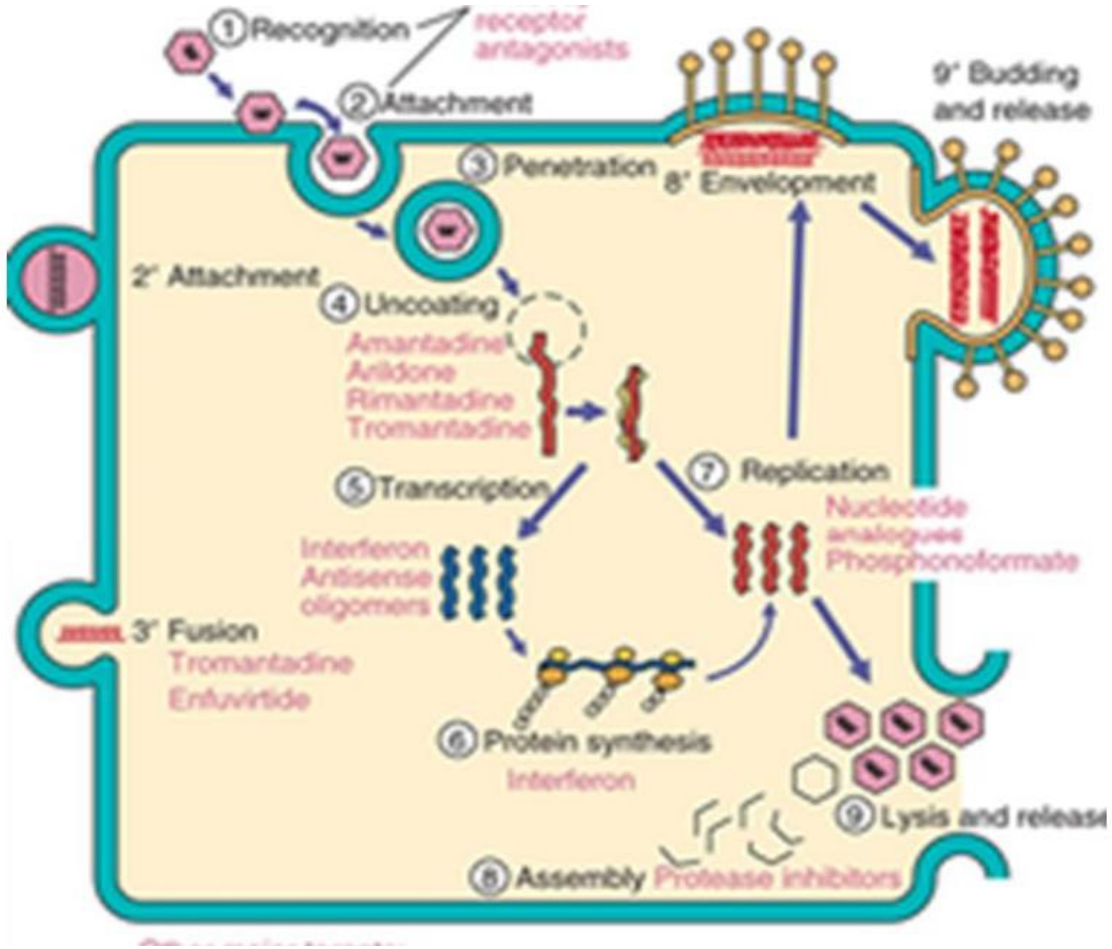
- **Arboviruses include:**
- **Toqaviridae,**
- **Flaviviridae,**
- **Arenaviridae,**
- **Bunyaviridae**
- **Roboviruses include:**
- ***Bunyaviridae* (genus *Hantavirus*),**
- ***Arenoviridae***
- ***Filoviridae***

Togaviridae





Reproduction of togaviruses

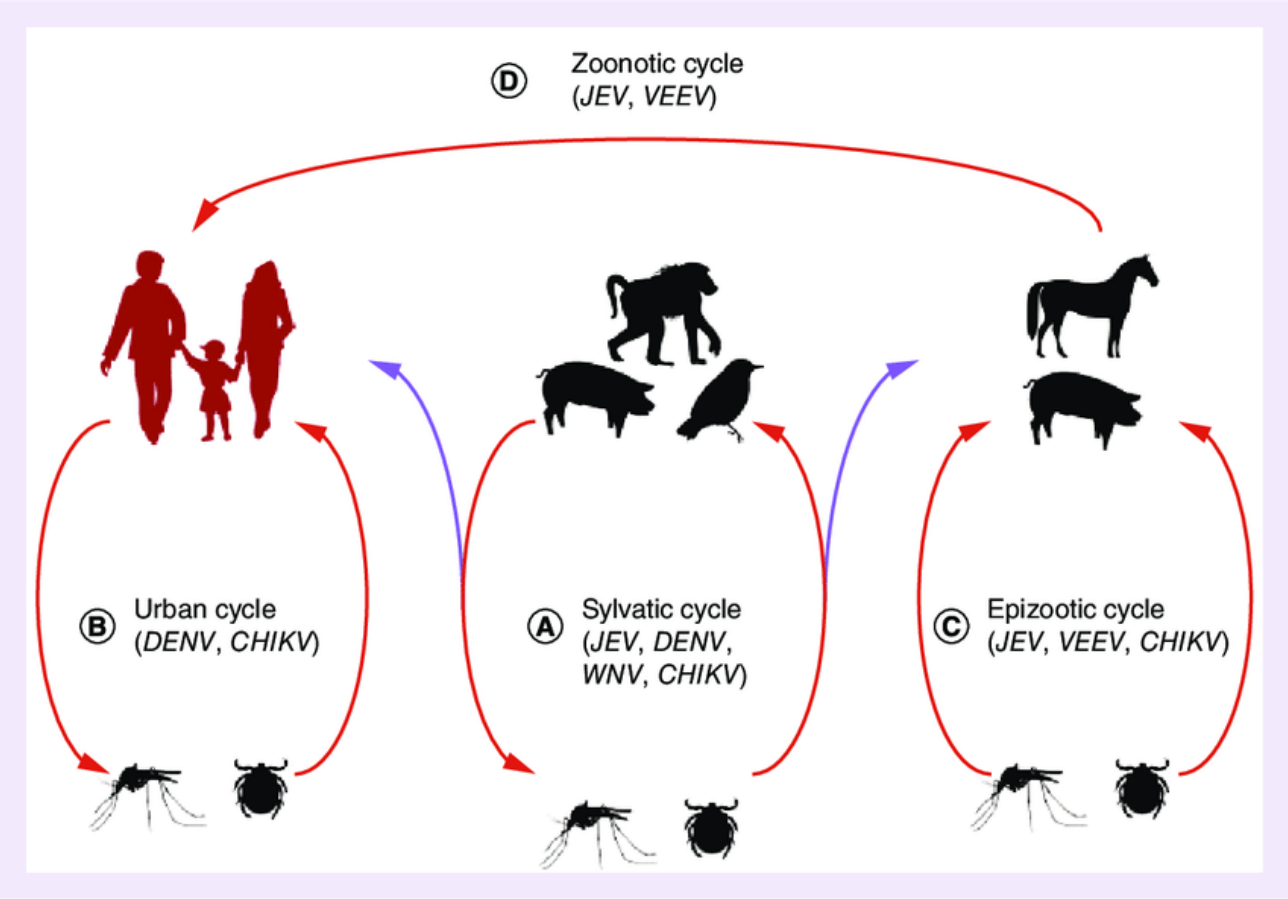


Classification of togaviruses

- 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.

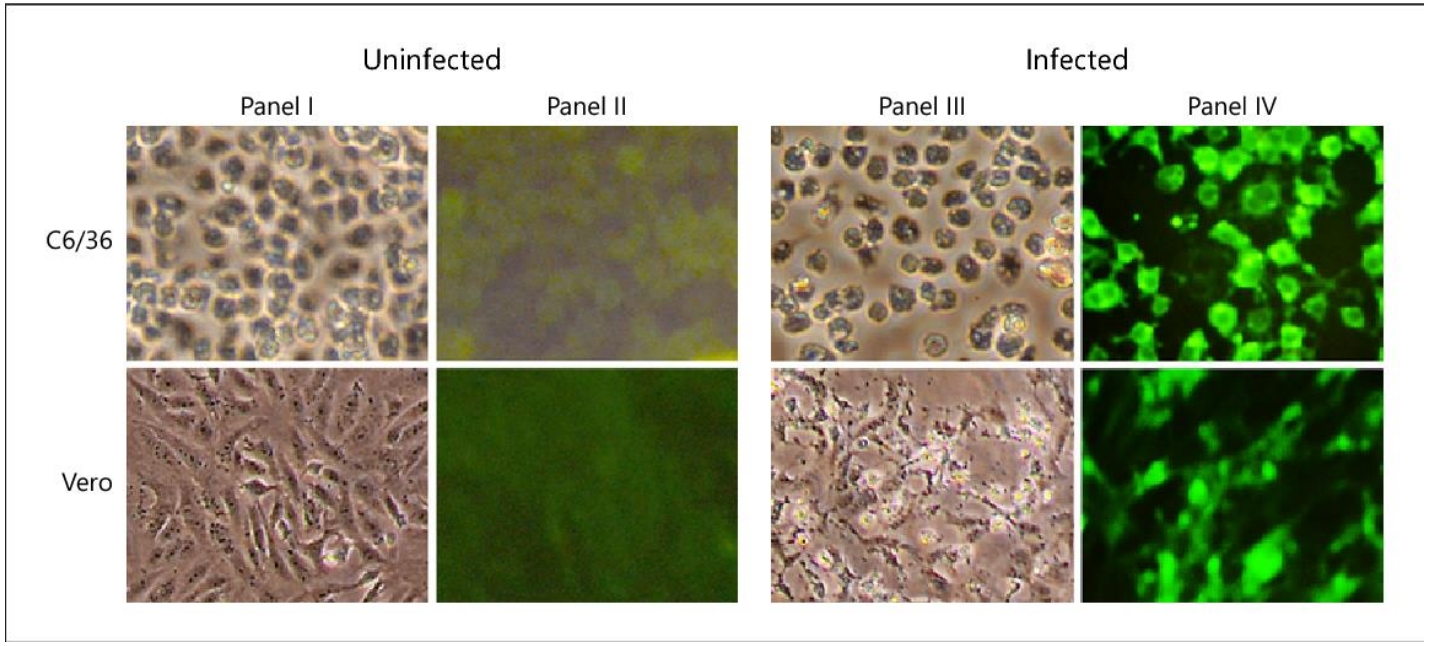
Clinical forms of alfavirus infections

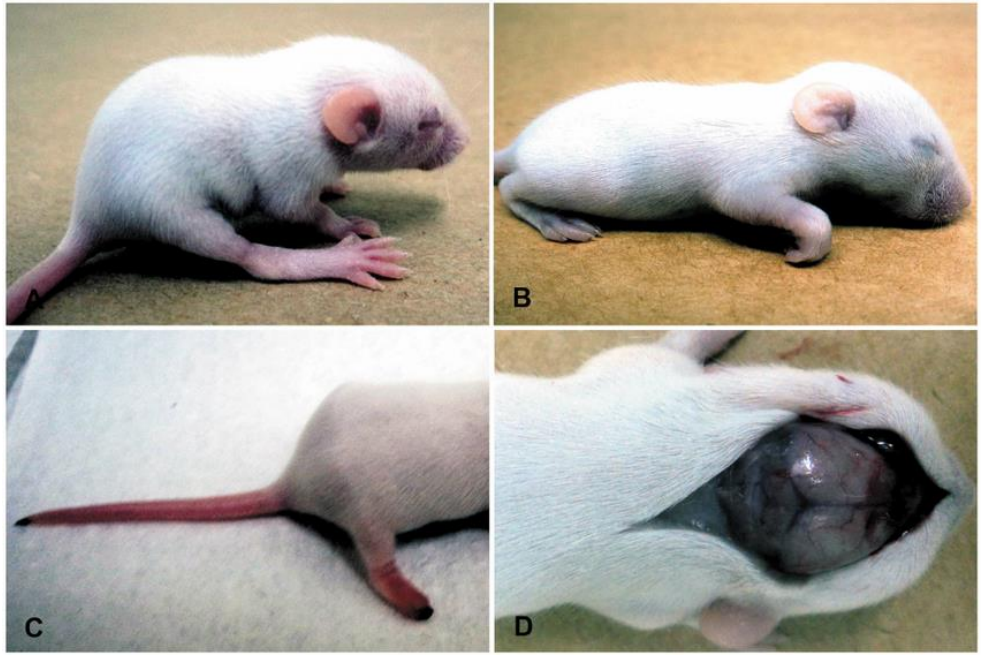
- ***Sindbis fever.*** The causative agent of the virus was obtained from mosquitoes in the village of Sindbis in Cairo (Egypt). The disease begins with fever, headache, arthralgia, skin rash and lasts 5-8 days. Although it has a benign end, it can become chronic and disability with the development of 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 alfavirus 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 serobrospinal 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 alfavirus infections





Flaviviridae

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

Flaviviruses

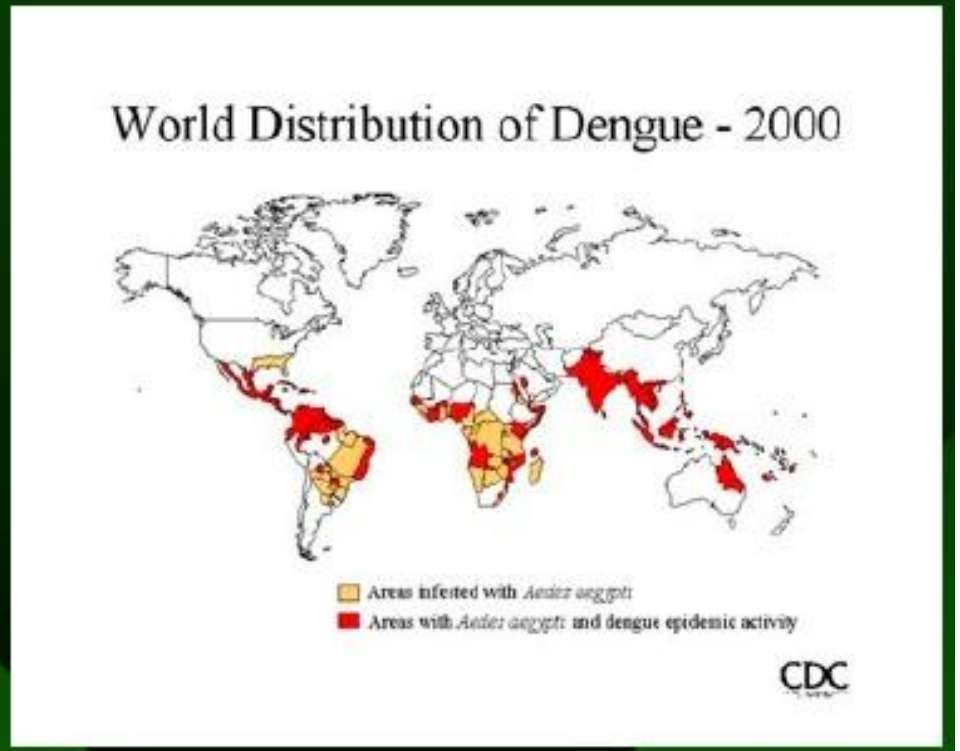
- Yellow Fever virus
- Dengue viruses
- St. Louis encephalitis virus
- Japanese encephalitis virus
- West Nile virus
- Murray Valley encephalitis virus, tick-borne encephalitis viruses and others

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)



Aedes aegypti

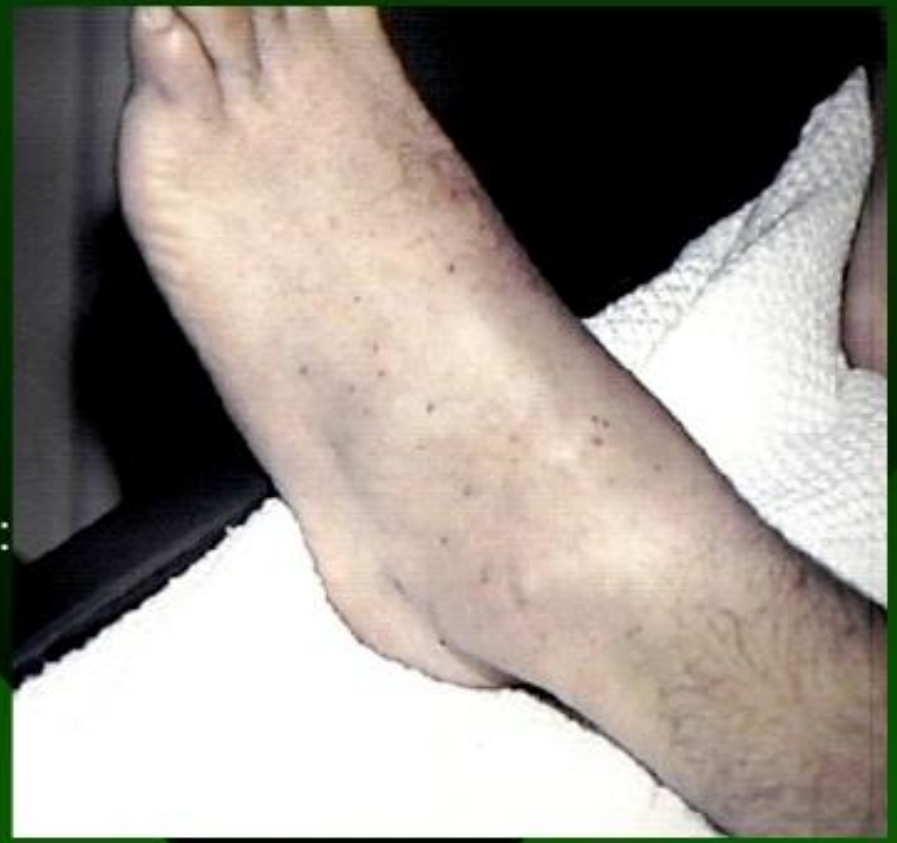
- Dengue transmitted by infected female mosquito
- Primarily a daytime feeder
- Lives around human habitation
- Lays eggs and produces larvae preferentially in artificial containers with clean stagnant water

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

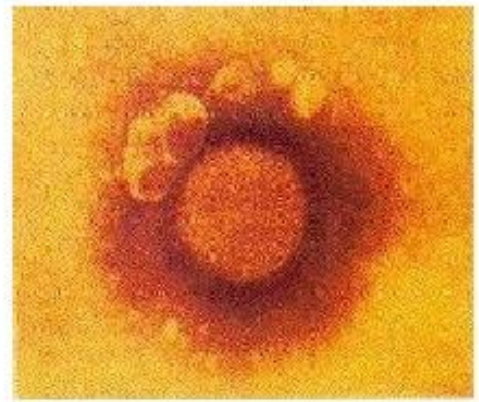
Japanese encephalitis virus

- 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)

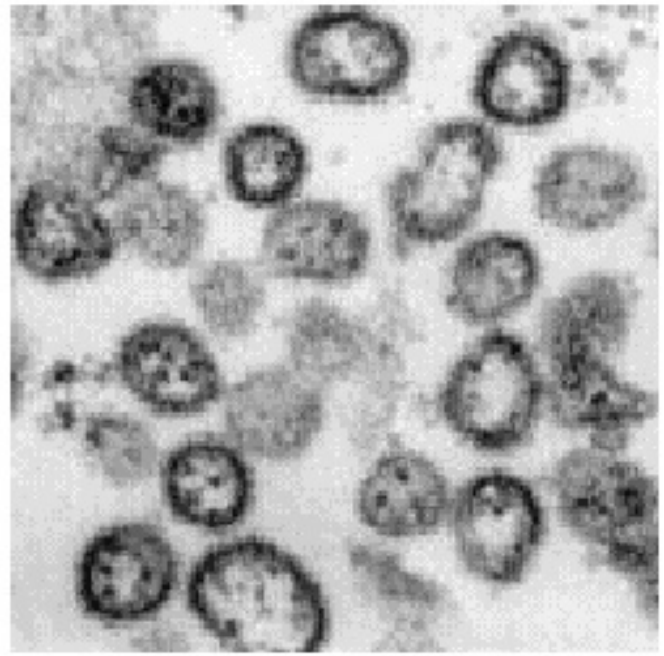
Arenaviruses



Introduction

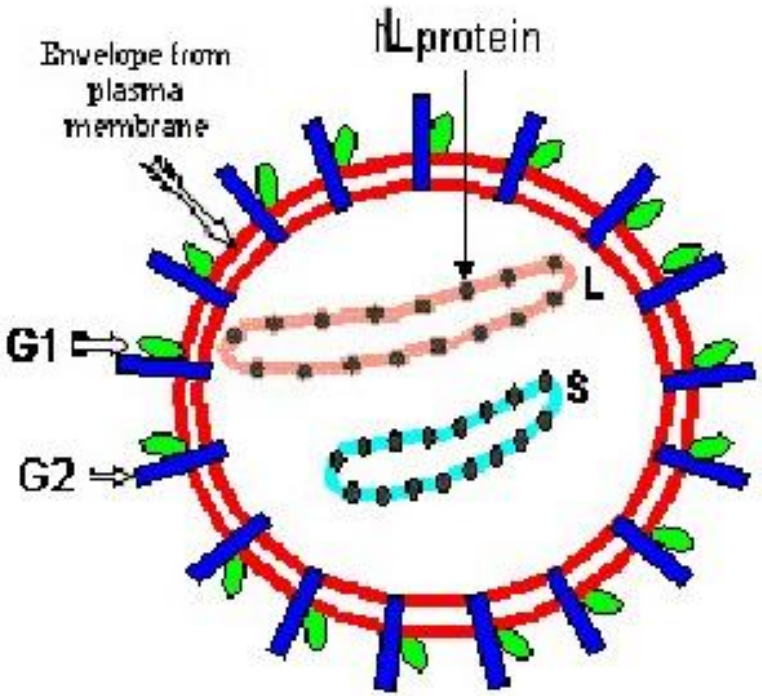
- Family = Arenaviridae
- Genus = Arenavirus
- Currently 22 recognized species
- 2 groups

- 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



Arenavirus particle

- 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 secreta

- inoculation via cuts, bites

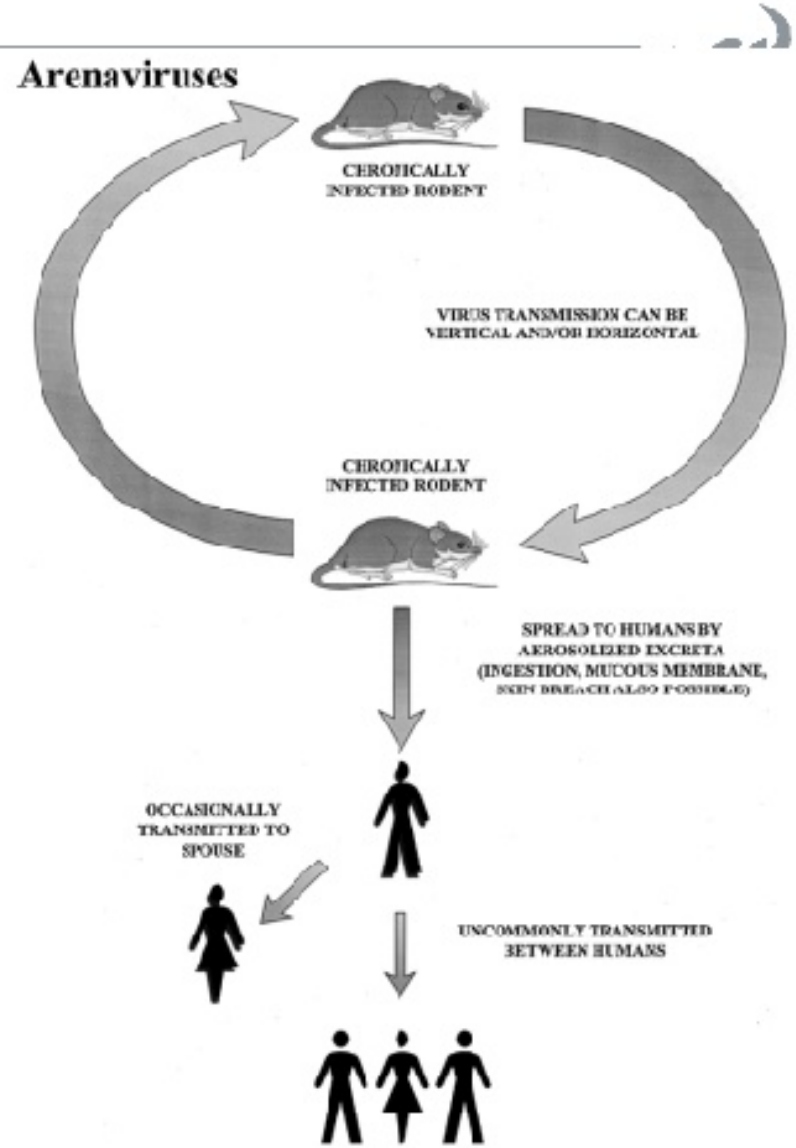
- contaminated fomites, food

-Rodent consumption

- Human – Human

- contaminated secretions, sexual

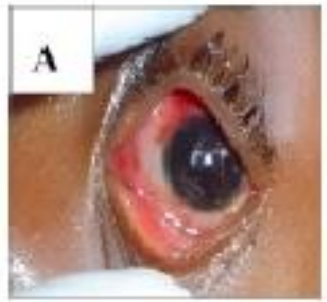
-Inoculation

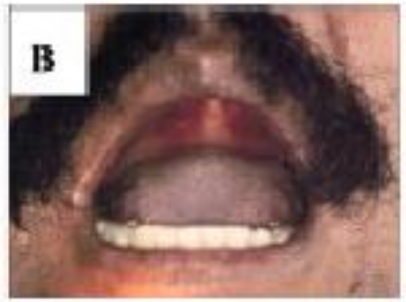


Clinical presentation

Table 2 Clinical stages of Lassa fever (adapted from McCarthy 2002[11])

Stage	Symptoms
1 (days 1-3)	General weakness and malaise. High fever, >39°C, constant with peaks of 40-41°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

- SS RNA
C=122

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

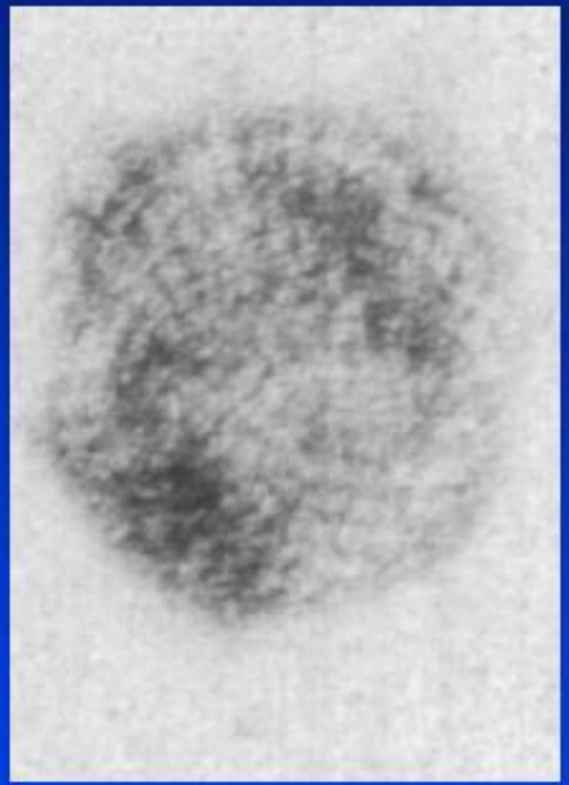
- Headache
- pain in limbs
- bleeding from multiple orifices

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

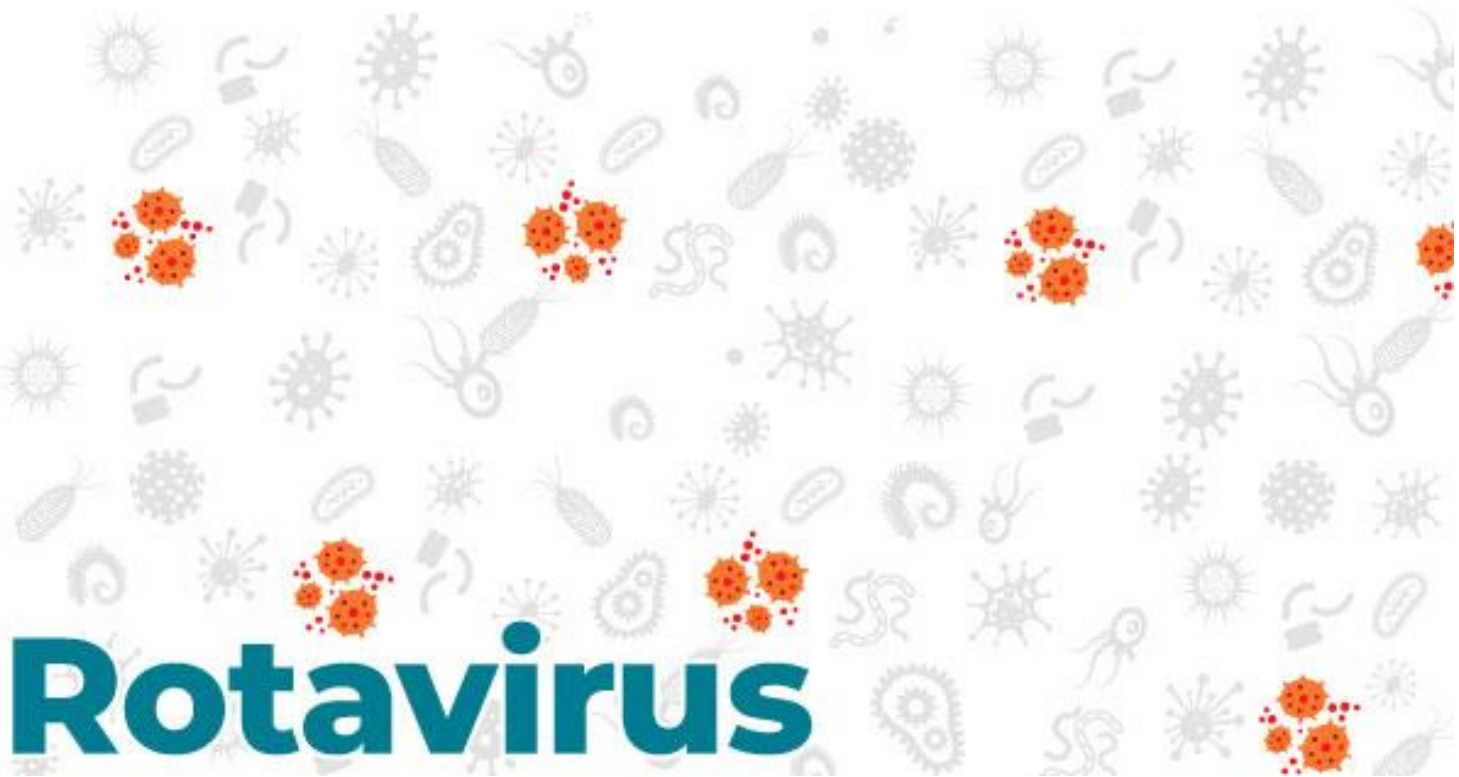


Hanta Virus

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

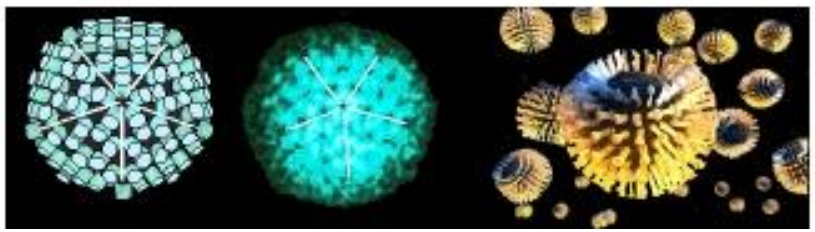
Hantavirus Pulmonary Syndrome

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


Rotavirus

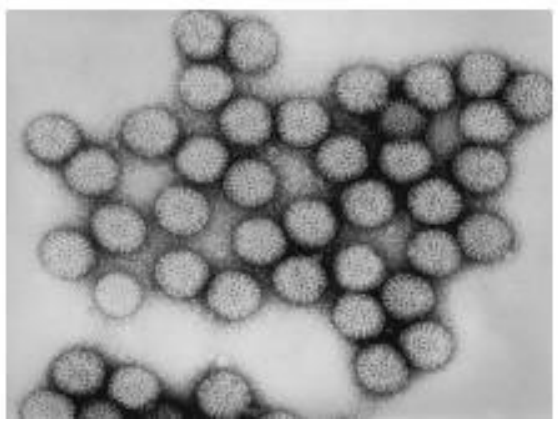
Rotavirus - Structural features

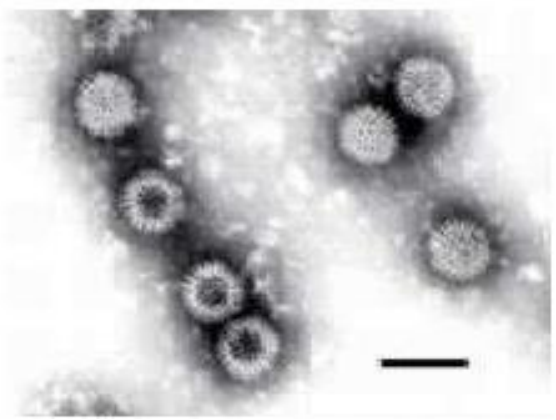
- Reovirus (RNA)
- 60-80nm in size
- Double stranded (ds) RNA
- Non-enveloped virus
- A rotavirus has a characteristic wheel-like appearance when viewed by electron microscopy
  - The name rotavirus is derived from Latin, meaning "wheel"
- Group A is important human pathogen [7 Groups (A to G)]
- 5 predominant strains (G1-G4, G9), account for 90% of isolates
- Strain G1 accounts for 73% of infections



Characters

- The virus is stable in the environment
- Relatively resistant to hand-washing agents
- Susceptible to disinfection
  - 95% ethanol, ‘Lysol’, formalin
- Very stable and may remain viable for weeks or months if not disinfected





Transmission

- Transmission
  - Mainly person to person via fecal-oral route, fomites
  - Poor hygiene
- Food and water-borne spread is possible
- Spread via respiratory route is speculated




Pathogenesis

- Reservoir — Human-GI tract
- Communicability — 2 days before to 10 days after onset

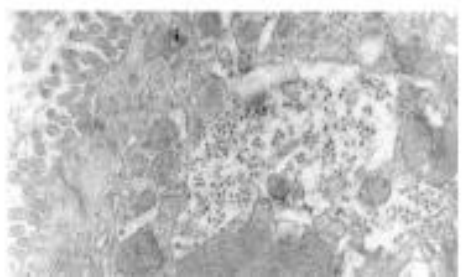
- Entry through mouth
- Replication in epithelium of small intestine
- Viremia uncommon
- Infection leads to isotonic diarrhea

Pathogenicity

- The virus infect the villi of the small intestine
  - Gastric and colonic mucosa are not infected
- Attach with the enterocytes by VP4
- They multiply in the cytoplasm of the enterocytes and damage their transport mechanisms
- Damaged cell may show into lumen of the intestine and release large quantities of virus which appear in the stool
- Viral excretion usually lasts for 2 – 12 days in otherwise healthy patients

Mechanism of diarrhea

- They strip the tips of the villi thus decreasing the surface area and decreasing by more than 50% the specific absorptive capacities of the intestine
- Damaged cells on villi are replaced by non absorbing immature cells
- Watery diarrhea due to net secretion of intestinal fluid and loss of absorptive surface
- Activation of the enteric nervous system
- Role of NSP4 peptide regions as an enterotoxin

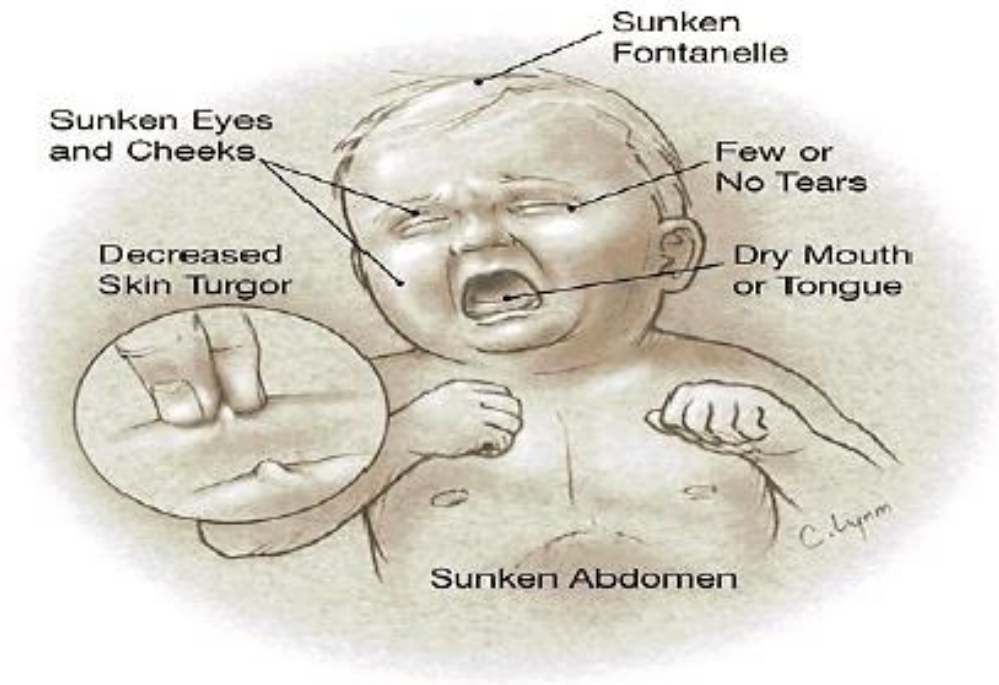


Clinical Features

- Incubation period 1-3 days
- Clinical manifestations depend on whether it is the first infection or reinfection
- Present with
  - Watery diarrhea (no blood or leukocytes)
  - Fever, can be high grade
  - Abdominal pain
  - Vomiting
  - Loss of electrolytes and fluids leading to dehydration
  - May be fatal unless treated

- First infection after age 3 months generally most severe
  - May be asymptomatic or result in severe dehydrating diarrhea with fever and vomiting
- GI symptoms generally resolve in 3 to 7 days

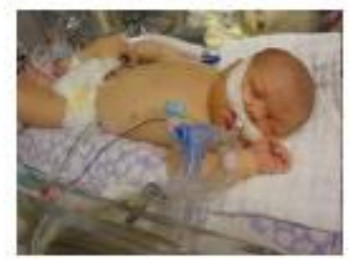
Dehydration - leading cause of morbidity and mortality



Complications

- Severe chronic diarrhea
- Dehydration
- Electrolyte imbalance
- Metabolic acidosis
- Immunodeficient children may have more severe or persistent disease



Immunity

- Antibody against VP7 and VP4, and Secretory IgA probably important for protection
- First infection usually severe
  - First infection usually does not lead to permanent immunity
  - Subsequent infections generally less severe
- Re-infection can occur at any age
- By age 3 years, 90% of the children have serum antibodies to one or more types
- Young children may suffer up to five re-infections by 2 years of age

Diagnosis

- Serology for epidemiologic studies
  - Antigen detection in stool
  - Antibody detection in serum
- Molecular methods
- Electron Microscopy
- Culture
  - Group A Rotaviruses can be cultured in monkey kidney cells
- Histopathology

Serology

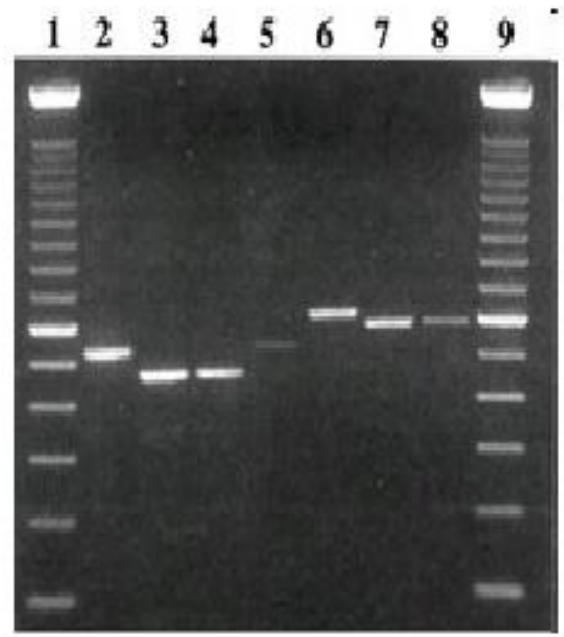
- Antigen detection in stool
  - ELISA, LA (Group A Rotavirus), ICT
- Antibody detection
  - ELISA can detect antibodies and establish rise in titers
- Serology for epidemiologic studies

Microscopy (EM)

- Demonstration of Virus in stool helps in early disease
- Electron Microscopy has made the identification simpler
- Non-Group A viruses also

PCR/Genotyping

- Genotyping is most sensitive method for detection of Rotavirus NA from stool specimens



Treatment

- Treatment of Gastroenteritis is supportive
- Correction of loss of water and electrolytes remain the goal treatment
- Failure for prompt correction of dehydration leads to
  - Acidosis
  - Shock
  - Death
- Lesser deaths if effective fluid replacement therapy is timely initiated

Fluid Replacement

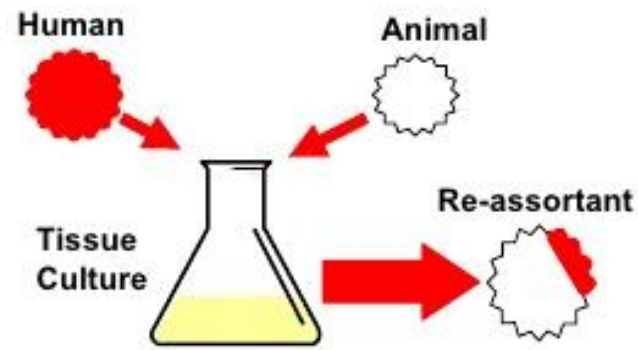
- Management consists of replacement of fluids (ORS) and restoration of Electrolyte balance
- Oral rehydration therapy is highly effective in reducing morbidity and mortality
- Severe dehydration needs parental administration of fluids

Prevention and Control

- In view of fecal-oral route of transmission, significant control measures are
  - Waste water management
  - Safe drinking water supplies
  - Sanitation
- Basic measures
  - Keep your hands clean
  - Wash hands often with soap and warm water after using the toilet, diapering and before preparing or eating food
- Vaccine

Vaccine

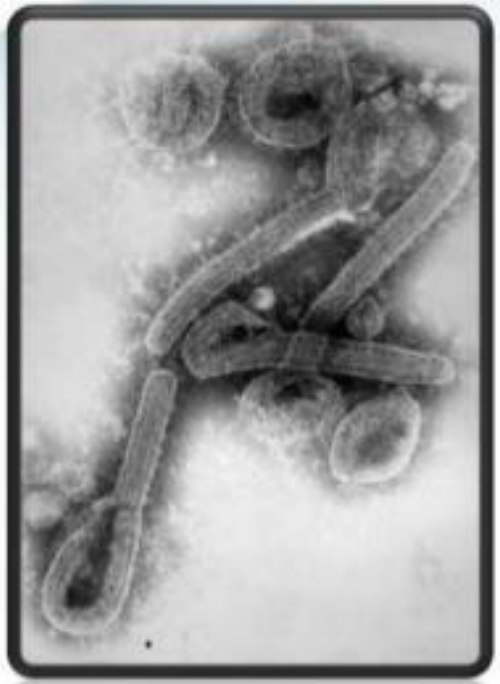
- A live, oral, pentavalent, human-bovine re-assortant vaccine
- Administered at 2, 4, and 6 months of age
  - RotaTeq™
  - Rotarix™



Created by genetic re-assortment of human and bovine antigens

FILOVIRUS

Ébola



Marburg

What is Ebola Virus?

- A notoriously deadly virus that causes fearsome symptoms
  - –High Fever
  - –Internal Bleeding
- kills as many as 90% of the people it infects

Introduction

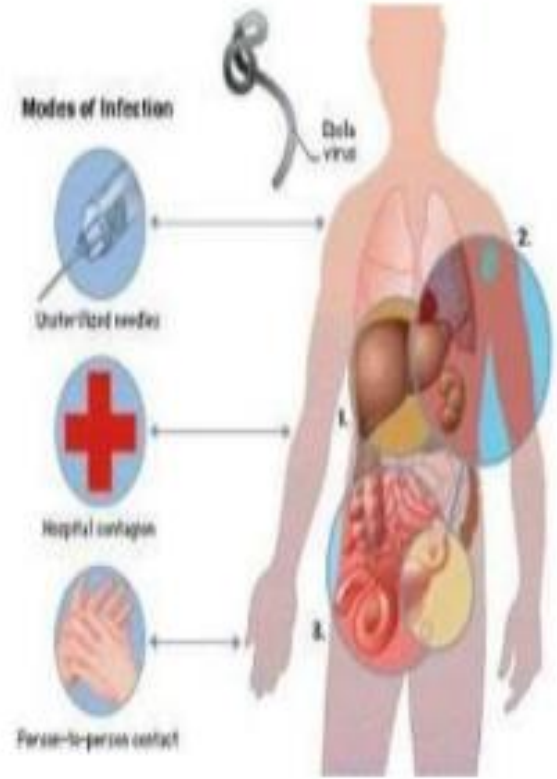
Ebola hemorrhagic fever (Ebola HF) is one of numerous Viral Hemorrhagic Fevers. It is a severe, often fatal disease in humans and nonhuman primates (such as monkeys, gorillas, and chimpanzees.

Structure

- Like all filoviruses, ebolavirions are filamentous particles shape of a shepherd's crook or in the shape of a "U" or a "6", and they may be coiled, toroid, or branched.
- In general, ebolavirions are 80 nm in width, but vary somewhat in length.
- In general, the median particle length of ebolaviruses ranges from 974 to 1,086 nm (in contrast to marburg virions, whose median particle length was measured at 795–828 nm), but particles as long as 14,000 nm have been detected in tissue culture.

MODE OF TRANSMISSION

- Unsterilized needles.
- Sub optical hospital conditions.
- Personal contact.
- Through blood to blood contact.
- Human to human transmission.
- Reusing needles and blood gloves in hospital.



Pathogenesis

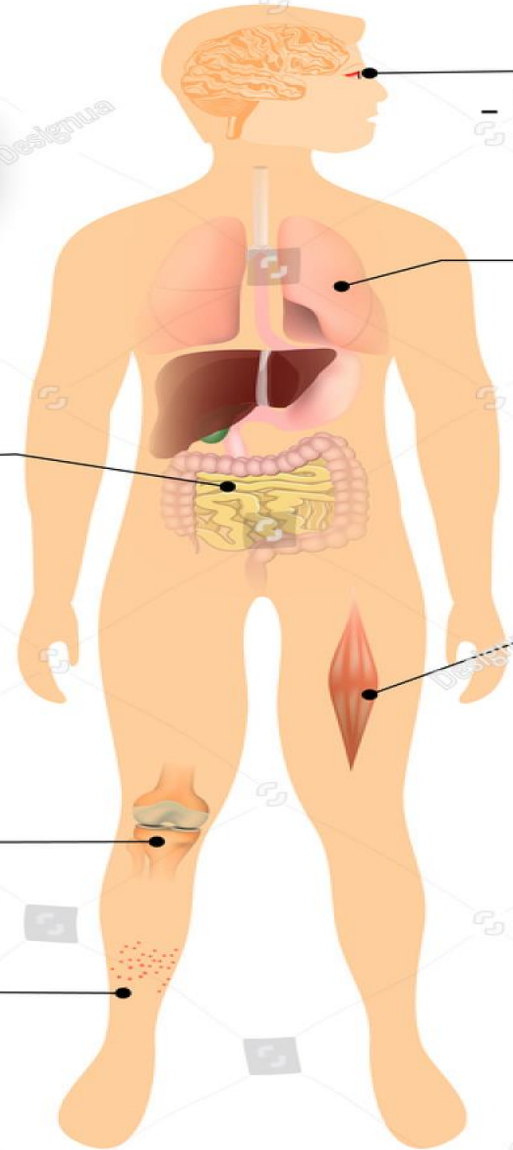
- Endothelial cells, mononuclear phagocytes and hepatocytes are the main targets of infection.
- After infection, a secreted glycoprotein (sGP) known as the Ebola virus glycoprotein (GP) is synthesized.
- Ebola replication overwhelms protein synthesis of infected cells and host immune defenses. The GP forms a trimeric complex, which binds the virus to the endothelial cells lining the interior surface of blood vessels.
- The sGP forms a dimeric protein that interferes with the signaling of neutrophils,which allows the virus to evade the immune system by inhibiting early steps of neutrophil activation.

Pathogenesis

- These white blood cells also serve as carriers to transport the virus throughout the entire body to places such as the lymph nodes, liver, lungs, and spleen.
- The presence of viral particles and cell damage resulting from budding causes the release of cytokines (to be specific, TNF-α, IL-6, IL-8, etc.), which are the signaling molecules for fever and inflammation.
- The cytopathic effect, from infection in the endothelial cells, results in a loss of vascular integrity. This loss in vascular integrity is furthered with synthesis of GP, which reduces specific integrins responsible for cell adhesion to the inter-cellular structure, and damage to the liver, which leads to coagulopathy.

Symptoms of Ebola

Ebola virus



Eye
- bleeding

Respiratory system
- sore throat
- chest pain

Digestive system
- nausea
- diarrhea
- vomiting
- abdominal pain

Muscle
- pain

Joints
- pain

Skin
- rashes

Fever
Headache
Weakness
Internal and external bleeding

Signs and symptoms

- Signs and symptoms of Ebola usually begin suddenly with am influenza-like stage characterized by fatigue, fever, headaches, joint, muscle and abdominal pain.
- Vomiting, diarrhea and loss of appetite are also common.
- Less common symptoms include: sore throat, chest pain, hiccups, shortness of breath and trouble swallowing.
- The average time between contracting the infection and the start of symptoms is 8 to 10 days, but it can vary between 2 and 21 days.
- Skin manifestations may include a maculopapular rash (in about 50% of cases).
- Early symptoms of EVD may be similar to those of malaria, dengue fever or other tropical fevers, before the disease progresses to the bleeding phase.

Signs and symptoms

- In 40–50% of cases, bleeding from puncture sites and mucous membranes (e.g. gastrointestinal tract, nose, vagina and gums) has been reported.
- In the bleeding phase, which typically starts 5 to 7 days after first symptoms internal and subcutaneous bleeding may present itself through reddening of the eyes and bloody vomit.
- Bleeding into the skin may create petechiae, purpura, ecchymoses and hematomas (especially around needle injection sites).
- Types of bleeding known to occur with Ebola virus disease include vomiting blood, coughing it up or blood in the stool. Heavy bleeding is rare and is usually confined to the gastrointestinal tract.
- In general, the development of bleeding symptoms often indicates a worse prognosis and this blood loss can result in death. All people infected show some symptoms of circulatory system involvement, including impaired blood clotting.
- If the infected person does not recover, death due to multiple organ dysfunction syndrome occurs within 7 to 16 days (usually between days 8 and 9) after first symptoms.

Laboratory diagnosis

Laboratory tests used in diagnosis include:

Timeline of Infection	Diagnostic tests available
Within a few days after symptoms begin	- Antigen-capture enzyme-linked immunosorbent assay (ELISA) testing - IgM ELISA - Polymerase chain reaction (PCR) - Virus isolation
Later in disease course or after recovery	- IgM and IgG antibodies
Retrospectively in deceased patients	- Immunohistochemistry testing - PCR - Virus isolation

Prevention

Quarantine

- Quarantine, also known as enforced isolation, is usually effective in decreasing spread.
- Governments often quarantine areas where the disease is occurring or individuals who may be infected. In the United States, the law allows quarantine of those infected with Ebola.The lack of roads and transportation may help slow the disease in Africa. During the 2014 outbreak, Liberia closed schools.[

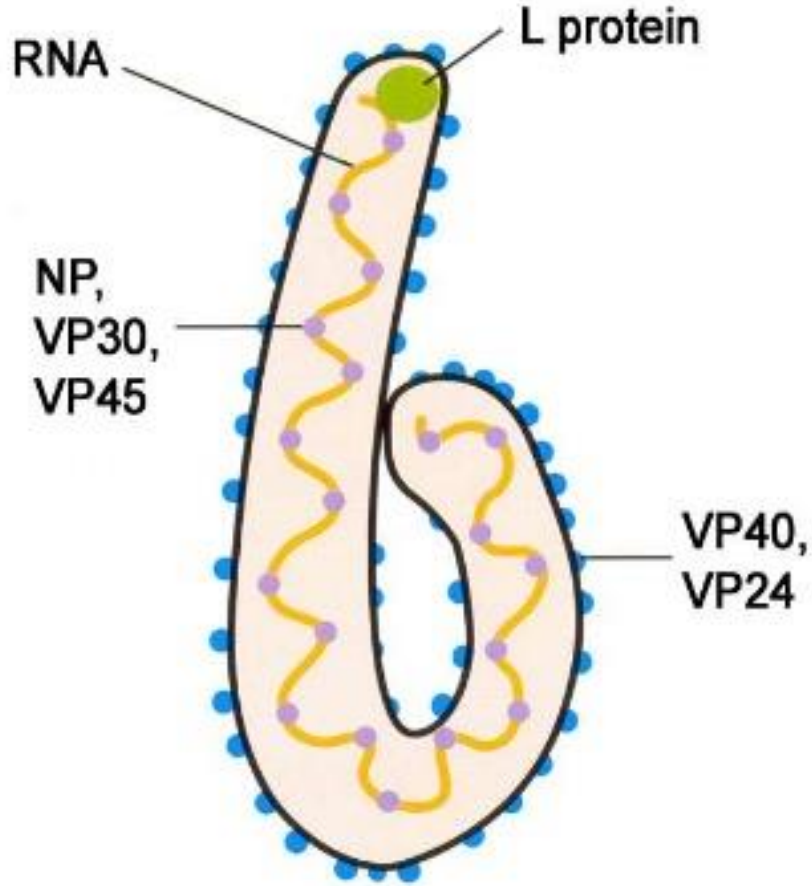
Vaccine

- No vaccine is currently available for humans.
- The most promising candidates are DNA vaccines or vaccines derived from adenoviruses, vesicular stomatitis Indiana virus (VSIV) or filovirus-like particles (VLPs) because these candidates could protect nonhuman primates from ebolavirus-induced disease. DNA vaccines, adenovirus-based vaccines, and VSIV-based vaccines have entered clinical trials

General Characteristics

- Order : Mononegavirales
- Family : Filoviradae
- Genus : Marburgvirus
- Species : Marburg marburgvirus

- Synonyms : Marburg disease, Marburg hemorrhagic fever, African hemorrhagic fever, and green monkey disease.



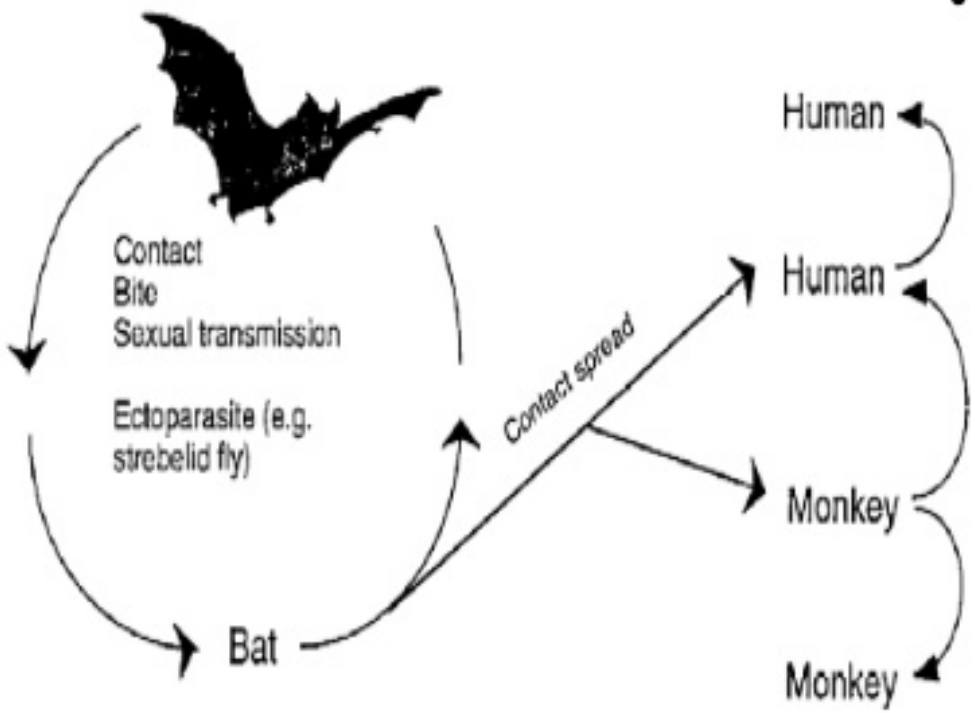
(it has the same structural properties as the Ebola virus)

Morphology

- Marburg is an enveloped, single-stranded, unsegmented, negative-sense RNA virus.
- It has the filamentous structure, can appear shaped like a U, a 6, or spiraled like a snail; and can sometimes be branched.
- They tend to include long noncoding regions at their 3' and/or 5' ends, which probably contributes to the stability of the viral transcript.
- The viral fragment is pleomorphic.
- Complexed with the proteins NP, VP35, VP30, and L.

Vector

- The natural reservoir for the virus is unknown. Epidemiologists have tested bats, monkeys, spiders and ticks for the virus, but were not able to acquire definitive data. Common factors indicate that the natural reservoir is part of rural Africa.



- Secondary spread of the disease is via contact with infected persons or contact with blood, secretions, or excretions of infected persons. The virus may continue to be shed in the patient's semen for up to 3-4 months after illness. Sexual transmission of the disease did occur in one instance in Germany.

PATHOGENICITY/TOXICITY

- A rare, severe hemorrhagic fever in humans and non-human primates characterized by a sudden onset with high fever, chills, headache, myalgia, and maculopapular rash, possibly followed by vomiting, chest pain, sore throat, abdominal pain, and diarrhea.

- Symptoms become increasingly severe and may include inflammation of the pancreas, jaundice, severe weight loss, delirium, shock, liver failure, massive hemorrhage, and multi-organ dysfunction. Marburg disease has a fatality rate of approximately 25 %.

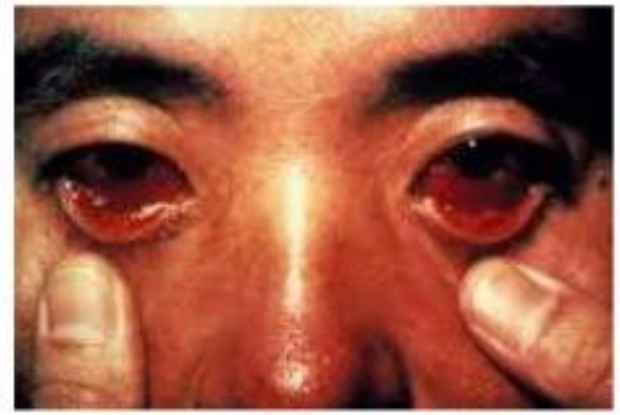
Mechanism

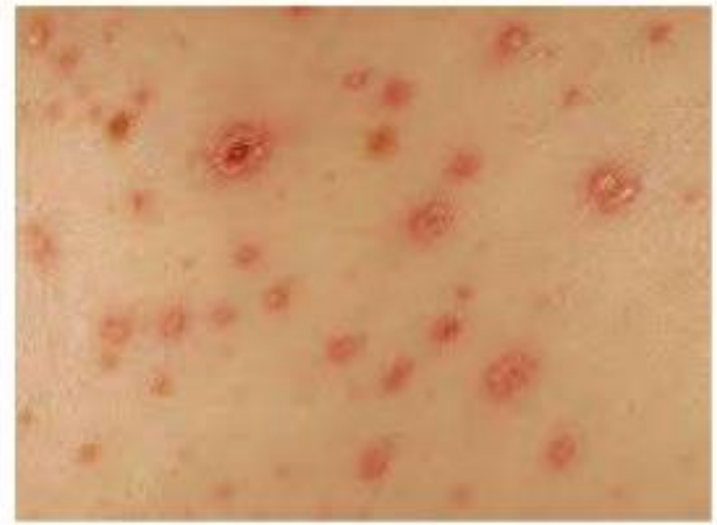
- As with Ebola, the exact mechanism of Marburg is unknown. However, virion surface spikes are made soley of large glycoprotein. It is presumed that, as with other negative-strand RNA viruses, these surface spikes bind to receptors on the host cell and mediate entry into susceptible cells.

- The Marburg virus has 22 potential *N*-linked glycosylation sites on its surface. Viral replication takes place in the cytoplasm, and envelopment is the result of budding preformed nucleocapsids. Systemically, the virus involves the liver, lymphoid organs, and kidneys.

- *Incubation period* : Usually 5-7 days, but can range from 3-10 days.

Symptoms





- Fever / Severe headache
- Joint and muscle aches
- Chills /Weakness
- Nausea and vomiting.
- Diarrhea (may be bloody)
- Red eyes.
- Raised rash.
- Chest pain and cough.
- Stomach pain.
- Severe weight loss.
- Bleeding, usually from the eyes, and bruising (people near death may bleed from other orifices, such as ears, nose and rectum)

Diagnosis

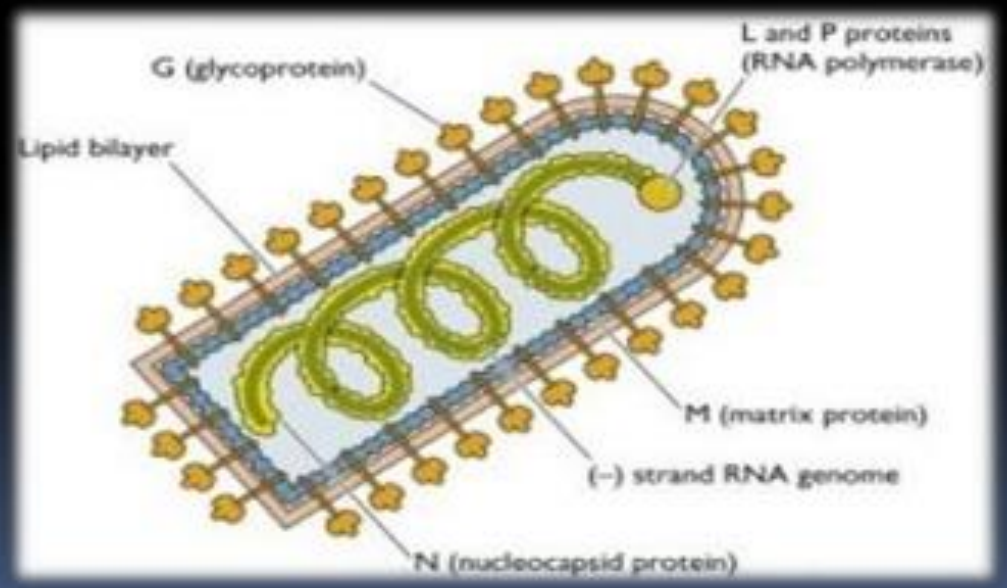
- For patients presenting with Marburg symptoms, initial possible diagnoses can include malaria and tyhpoid fever.
- As with Ebola, diagnosis of Marburg virus is confirmed by IgG ELISA, although IgM ELISA can be used to distinguish acute infections from old infections. IFA results can be misleading.
- Electron microscopy is useful in diagnosing filovirus infection, but does not help distinguish Marburg from Ebola.

Treatment & prevention

- Supportive therapy (there is no specific treatment for Marburg hemorrhagic fever. However, the virus itself is sensitive to lipid solvents, detergents, commercial hypochlorite disinfectants, and phenolic disinfectants. The virus can also be destroyed by ultraviolet and gamma radiation.

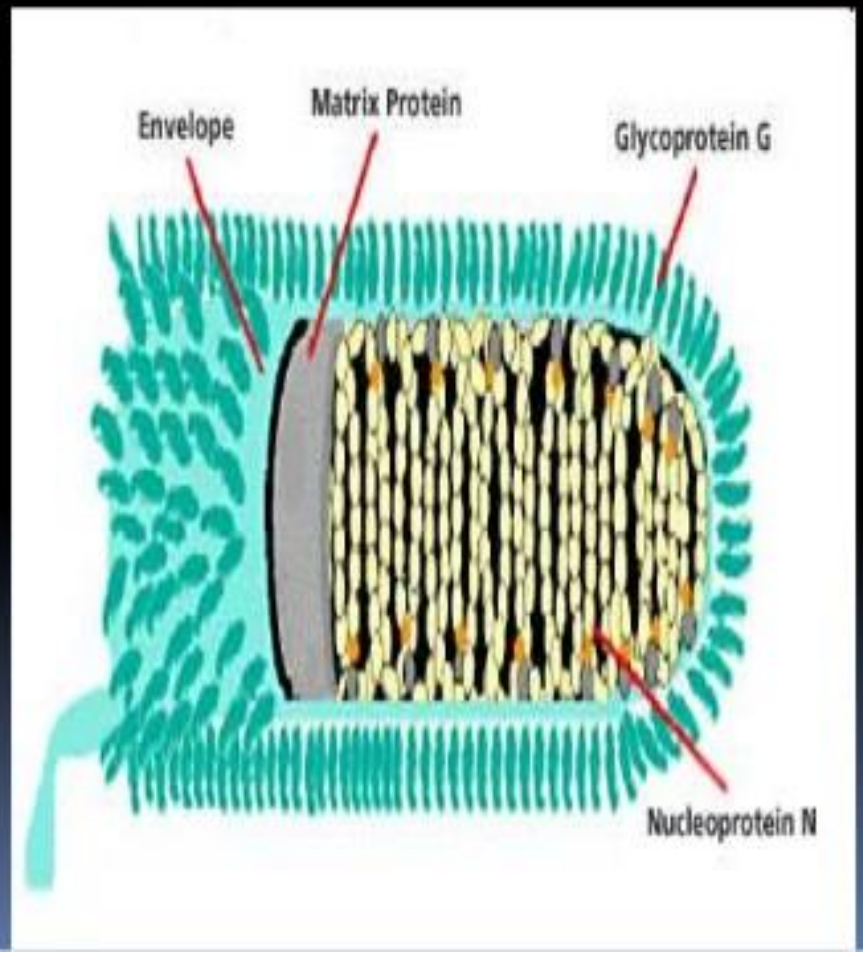
- Vaccine. None. As with exposure to other filoviruses, exposure to Marburg does not confer subsequent immunity. The antibody response in convalescent patients does not neutralize or protect against subsequent infection by Marburg virus.

RHABDO VIRUS

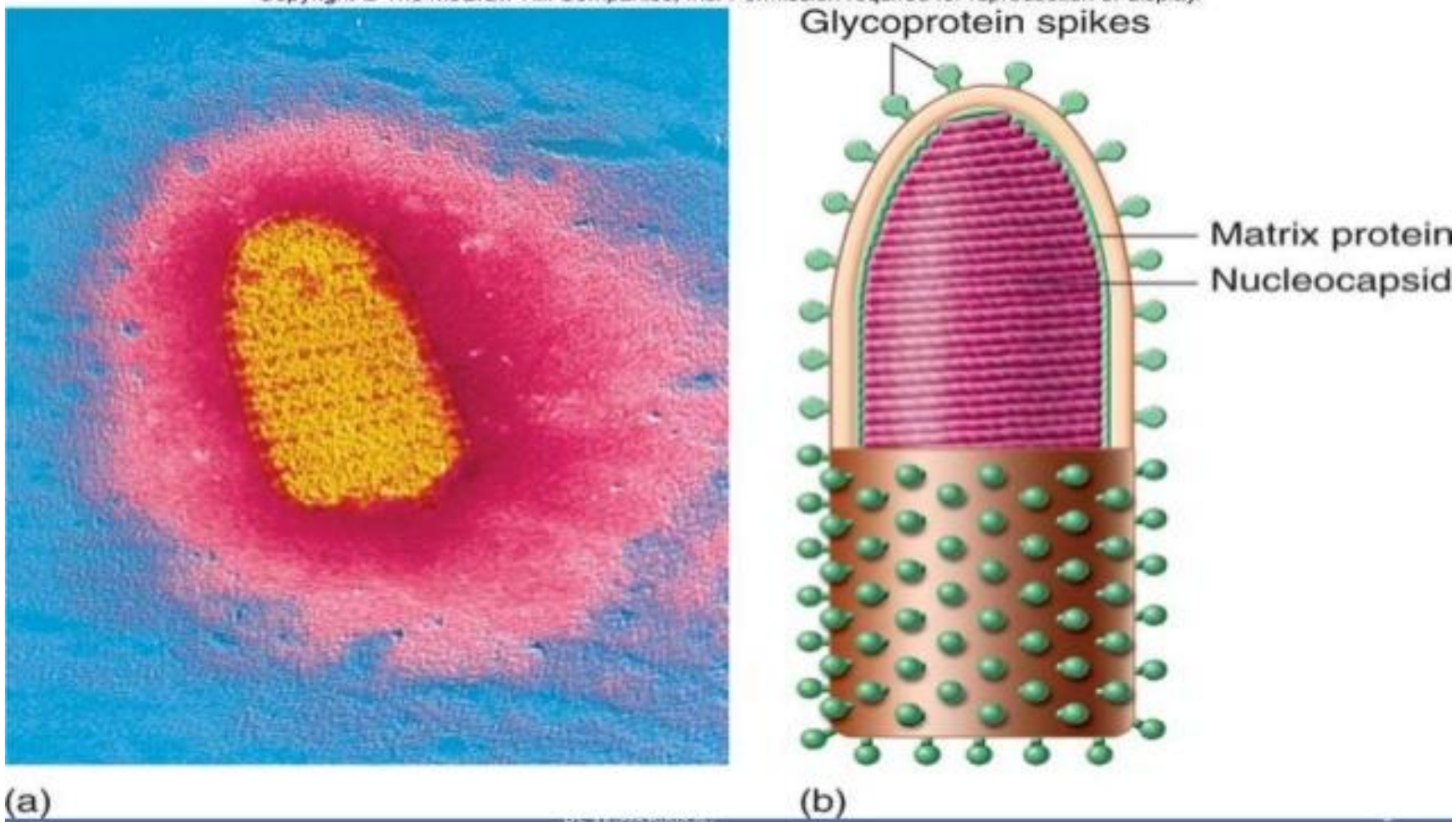


RhabdoVirus

- Single stranded ,linear ,negative sense ,non segmented RNA
- These are enveloped
- Bullet shaped virus
- Multiply in cytoplasm

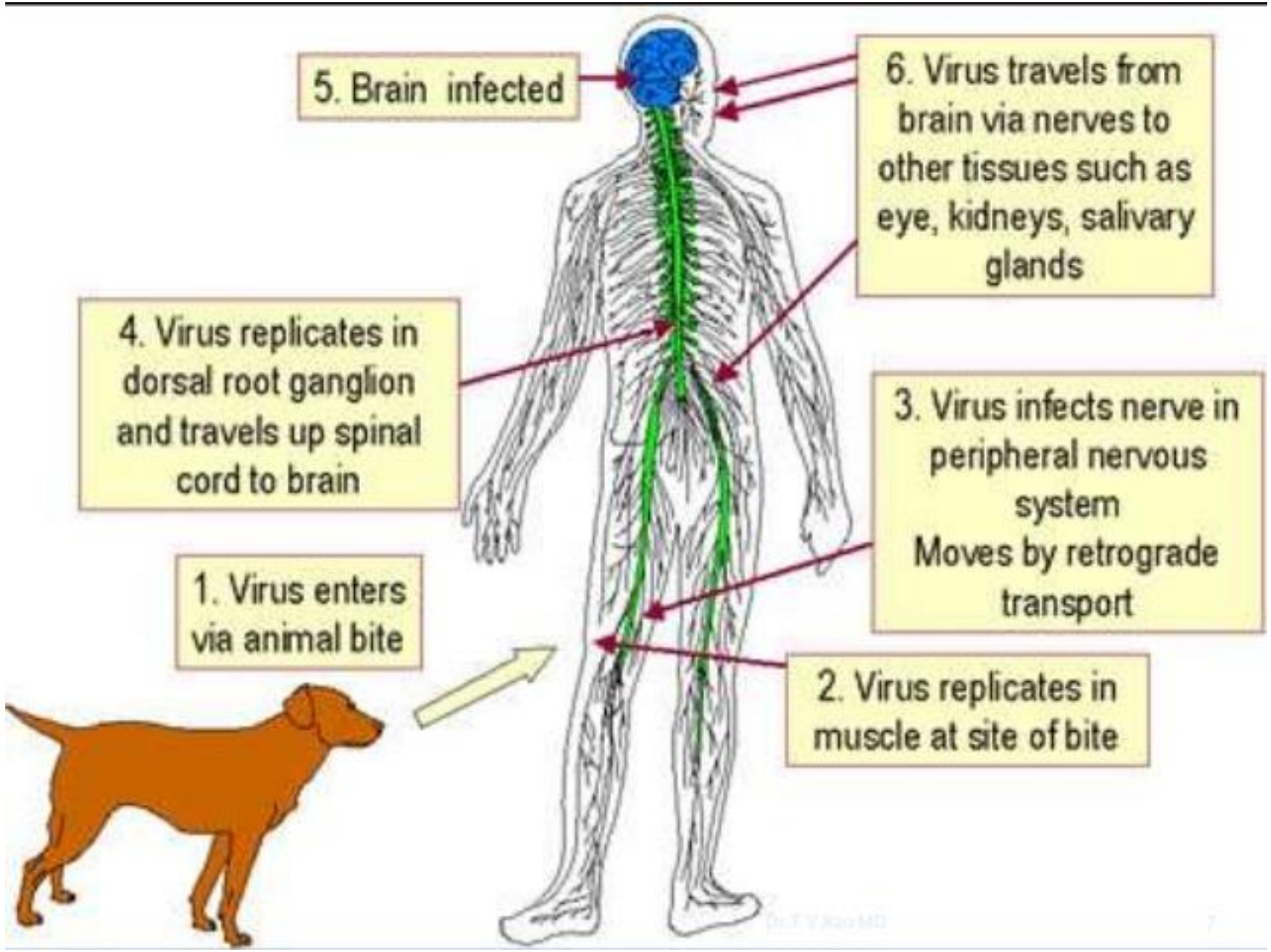


Structure of the rabies virus



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.


5. Brain infected
6. Virus travels from brain via nerves to other tissues such as eye, kidneys, salivary glands
4. Virus replicates in dorsal root ganglion and travels up spinal cord to brain
3. Virus infects nerve in peripheral nervous system
Moves by retrograde transport
1. Virus enters via animal bite
2. Virus replicates in muscle at site of bite

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 Findings

- Bizarre behavior.
- Agitation
- Seizures.
- Difficulty in drinking.
- Patients will be able to eat solids
- Afraid of water - Hydrophobia.
- Even sight of sound disturbs the patient.
- But suffer with intense thirst.
- Death in 1 -6 days.
- Respiratory arrest / Death / Some may survive.

Diagnosis

- Based on the history
- Signs and symptoms
- Clinical examination
- Detection of antigen by taking skin biopsy using immunofluorescence.
- Virus isolation from saliva & other secretions.
- CSF analysis, MRI and CT scan.
- ELISA
- RT-PCR
- direct Fluorescent Antibody (DFA) testing
- Negri bodies

Prevention

- Vaccination of susceptible animal species, particularly dogs and cats, will control this zoonotic disease.

Rabies PEP — Vaccination

- Previously unvaccinated persons get 4 doses
  - Days 0, 3, 7, and 14
  - 5th dose dropped from vaccine schedule last year
  - Intramuscular injections