

Lab 3:

Pathogenesis of Virus Infections & Pattern

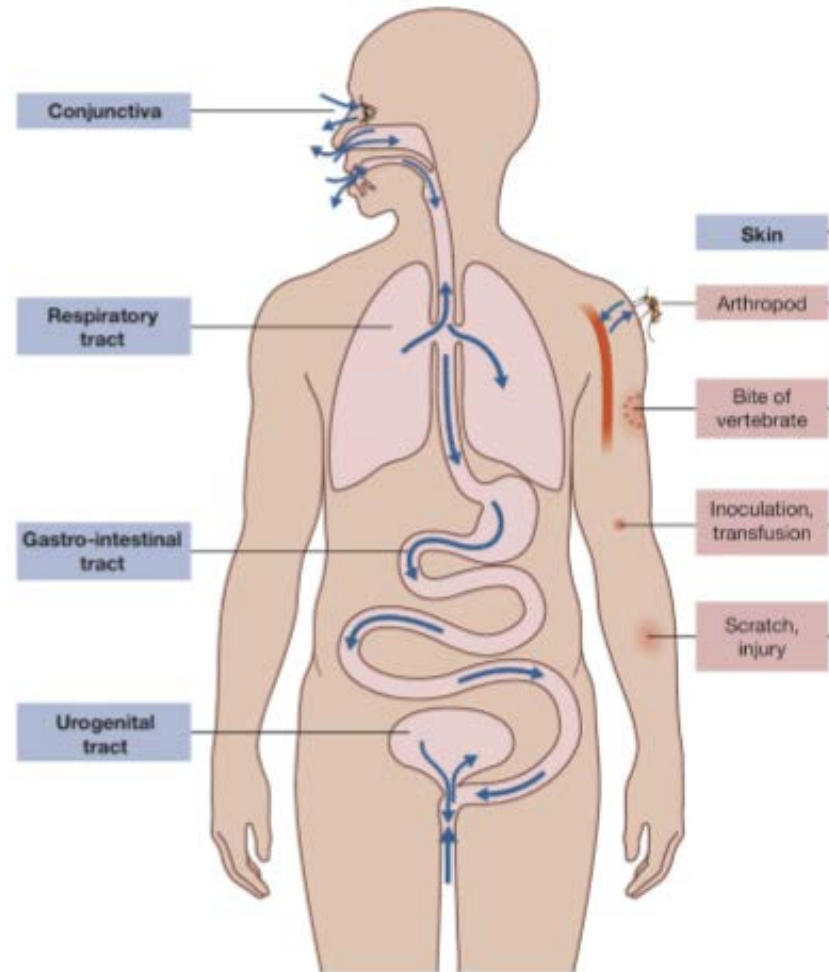
450 MIC

PRACTICAL PART
SECTION (30397)

Learning Outcomes

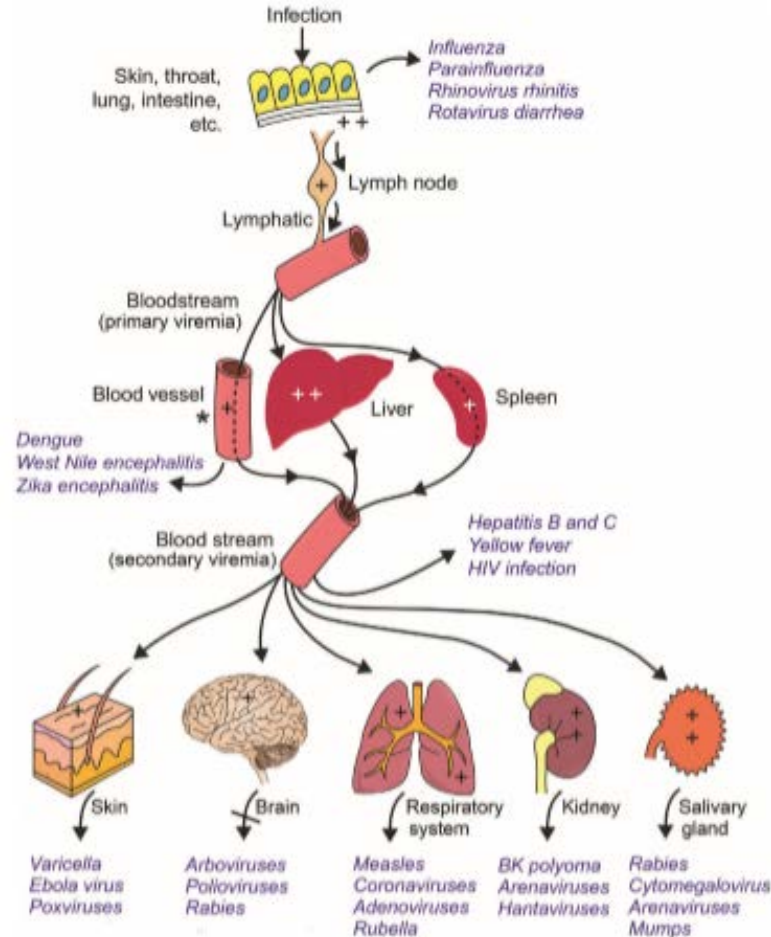
- The pathogenesis of viral infection
- The viral disease pattern
- Specific virus disease of human
- 1st: Herpes Simplex Virus (HSV)
 - Classification
 - Structure
 - Laboratory diagnosis

Figure 1: Routes of virus entry—the various surfaces of the body, one of which must be breached in order for infection to occur.



Reproduced from Flint, S.J., et al., 2009. Principles of Virology, third ed., Vol, II p. 348. ASM Press, Washington, DC, with permission.

Figure 2: The role of primary and secondary viremias in the spread of viruses throughout the body, showing some, sites of replication and routes of shedding. + indicates major sites of virus replication.



VIRUS INFECTION OF TARGET ORGANS

□ Different viruses present different unique patterns of infection (clinical signs, symptoms, laboratory data, etc.), based upon differences in their major sites of replication and damage “target organs.”

□ Other sites may also be infected at various stages of infection, without necessarily being clinically evident.

TABLE 7.5 Target Organs in Some Acute Transient Systemic Infections

Virus enters via the gastrointestinal tract

- Hepatitis A: liver
- Poliomyelitis: anterior horn cell in spinal cord and central nervous system
- Other enteroviruses: meninges, muscle, skin, CNS

Virus enters via the respiratory tract

- Chickenpox: (sensory ganglia), skin
- Measles: conjunctiva, skin, CNS
- Rubella: skin, joints
- Mumps: parotid and salivary glands, testes, pancreas, meninges
- Smallpox: skin, mucous membranes

What is Tropism?

The predilection of a particular virus for infecting a particular cell type or organ is known as its “tropism”.

With some viruses, the pathophysiological, molecular, and/or anatomical factors determining tropism have been partially clarified, but for many others these are not known.

For example: most viruses of humans replicate optimally at 37°C, while some respiratory viruses, as rhinoviruses replicate optimally at 33°C, (mucosal surfaces of the upper respiratory tract). Thus rhinovirus isolation is more successful if cell cultures are maintained at 33°C.





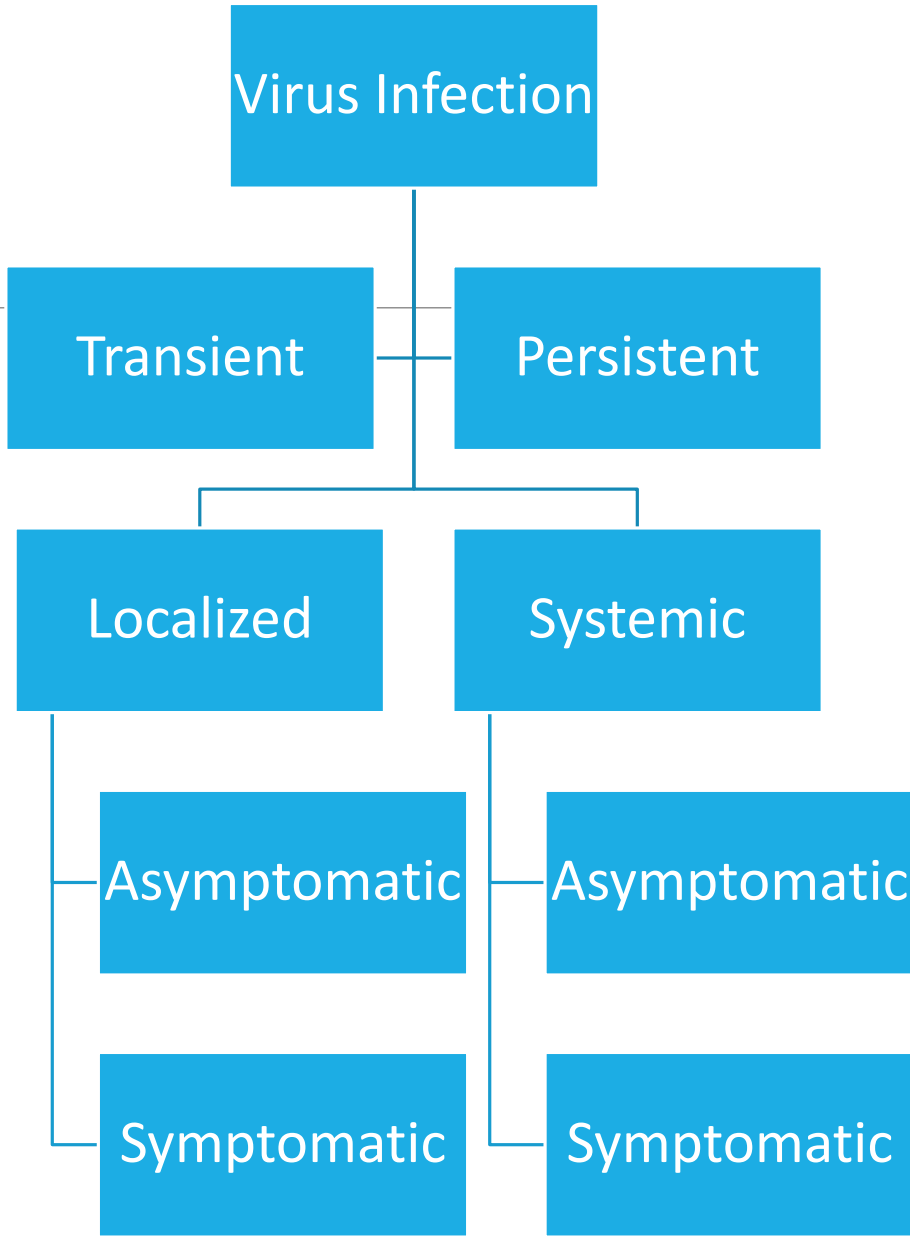
Patterns of Virus Infection

The four different infection models:

1- Transient localized, Transient systemic, Persistent localized, and Persistent systemic infections.

Different viruses typically cause specific patterns of disease and severity.

For example, 80 to 90% of measles and smallpox infections are symptomatic, whereas less than 5% of cytomegalovirus (CMV) infections are symptomatic.



Some Major Persistent Virus Infections of Humans

Virus	Site(s) of Persistence	Chronic Disease
Localized infection		
Papillomavirus	Skin, mucosal cells	Warts; genital and other cancers
Systemic infection		
<i>1- Acute infection with late rare complications</i>		
Measles	Central nervous system	SSPE (panencephalitis)
<i>2- Latent infection with reactivation</i>		
Herpes simplex 1 and 2	Sensory, autonomic ganglia	Cold sores, genital lesions
<i>3- Chronic infection with ongoing viral replication</i>		
Hepatitis B Virus	Liver, lymphocytes	Cirrhosis, liver cancer

Specimen Appropriate for Laboratory Diagnosis of Various Clinical Syndrome

Syndrome	Specimen
<i>Respiratory</i>	Nasal or throat swab; nasopharyngeal aspirate; sputum
<i>Enteric</i>	Feces
<i>Genital</i>	Genital swab, urine
<i>Eye</i>	Conjunctival (and/or corneal) swab
<i>Skin</i>	Vesicle fluid/swab/scraping; biopsy solid lesion
<i>Central Nervous System</i>	Cerebrospinal fluid; feces (enteroviruses)
<i>Generalized</i>	Throat swabs; feces; blood leukocytes
<i>Autopsy/Biopsy</i>	Relevant organ
<i>Any</i>	Blood

1st. Herpesviruses

Classification:

Order: Herpesvirales

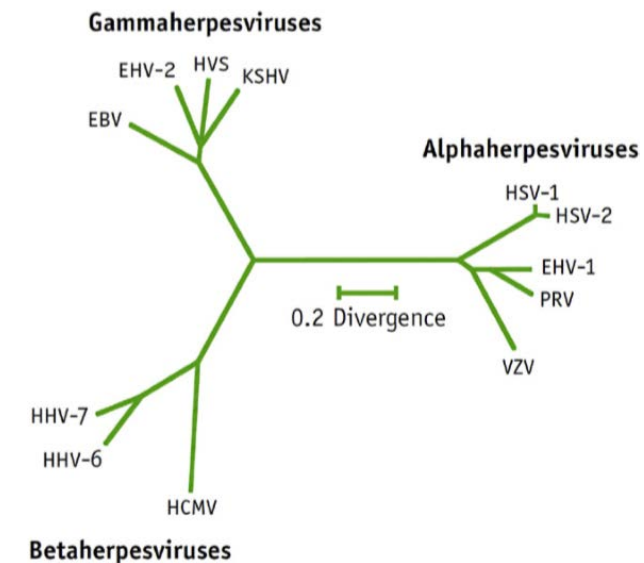
- **Families:** Herpesviridae

- **Subfamily1:** Alphaherpesvirinae

- human pathogens herpes simplex virus types 1 and 2

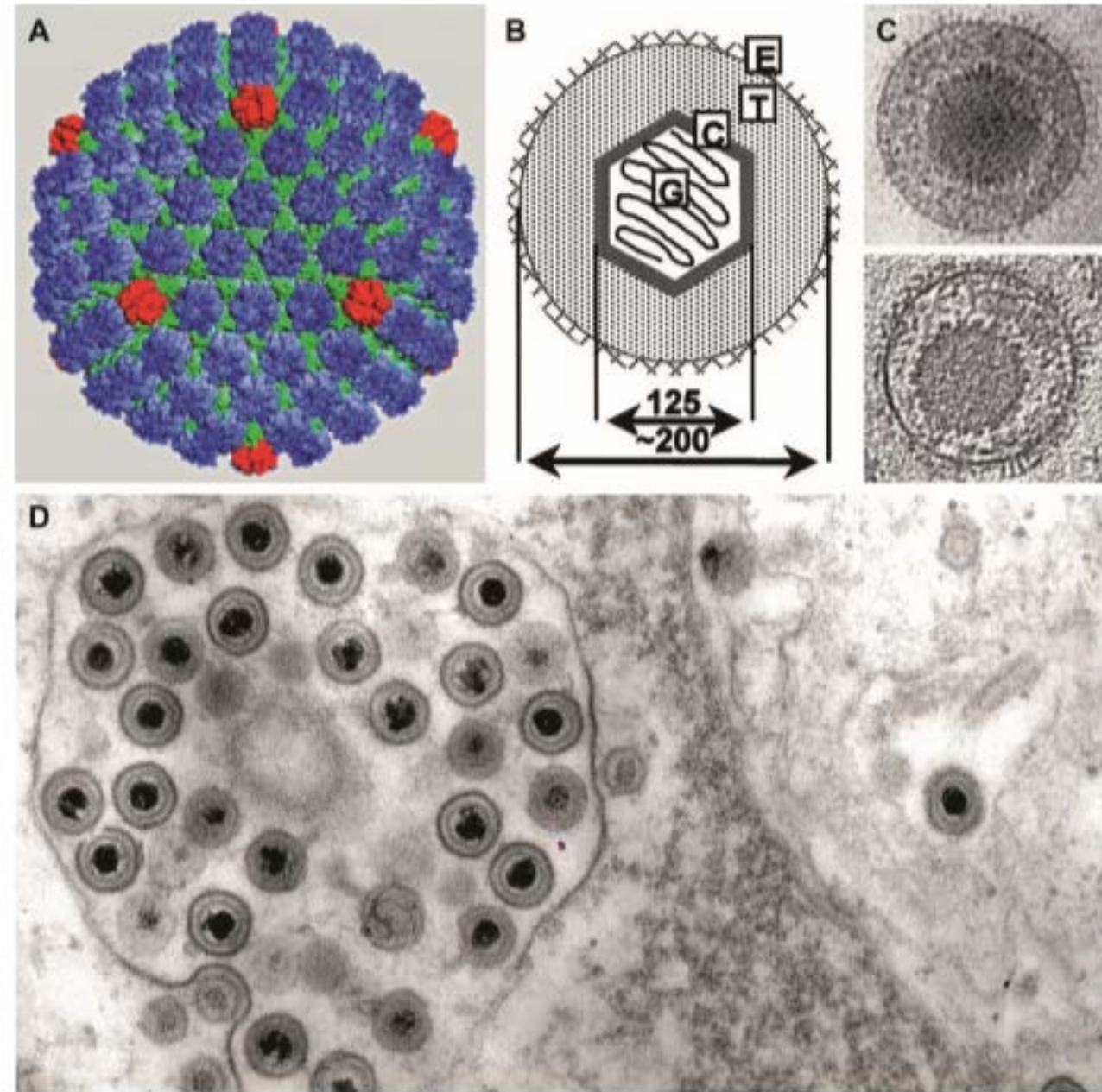
- **Subfamily2:** Betaherpesvirinae

- human cytomegalovirus, HHV-6 and HHV-7.



Herpesvirus (HSV) morphology and structure.

- (A) HSV type 1 virus icosahedral capsid.
- (B) Scheme of a HSV virion : G, genome; C, capsid; T, tegument; E, envelope.
- (C) Cryo-electron microscopy of HSV capsids.
- (D) Thin-section electron microscopy of HSV showing enveloped virions in various stages of maturation.



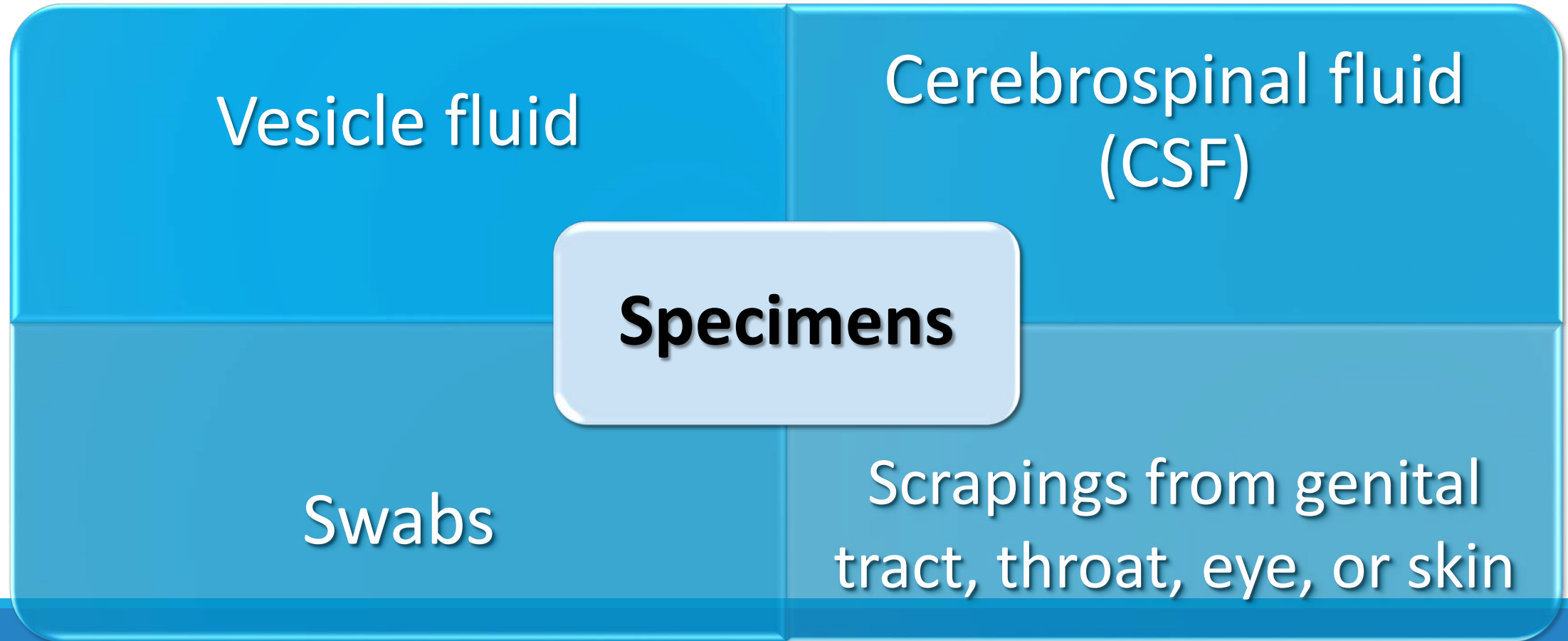
HERPES SIMPLEX VIRUS INFECTION

Herpes simplex virus infections of the lips, mouth, and genital tract were described in early Sumerian and Greek literature: the first successful, relatively non-toxic antiviral drug, acyclovir, was developed to treat herpes simplex



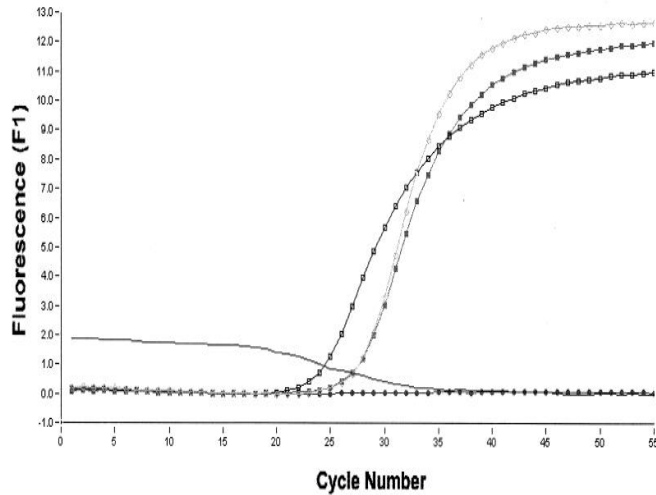
- (A) Lesions of recurrent labial HSV, in the healing and crusting phase.
- (B) Herpetic whitlow on the right thumb, occurring in a nurse.

Laboratory Diagnosis of Herpes Simplex Virus



For Cell Culture technique: the specimen should be taken early, placed in an appropriate transport medium, kept on ice, and transferred to a laboratory without delay.

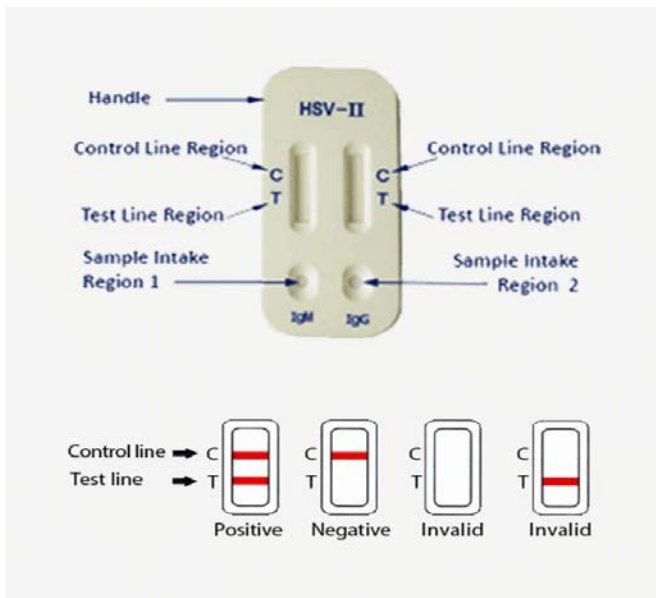
Laboratory Diagnosis of Herpes Simplex Virus



1-HSV DNA detection

By real-time polymerase chain reaction (PCR).

In detergent-solubilized cells or mucus from the site of lesions



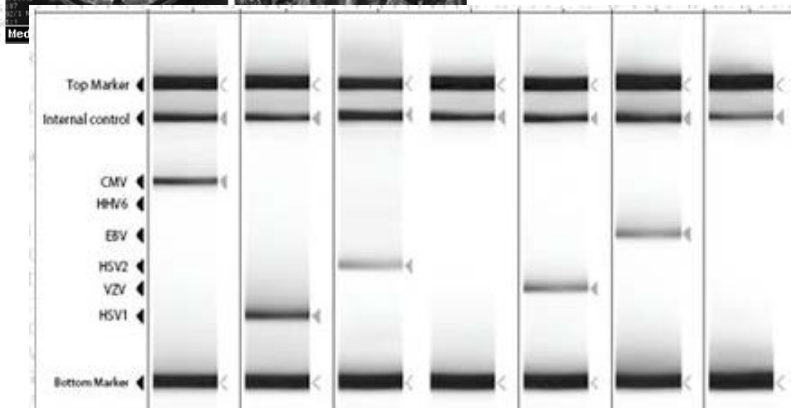
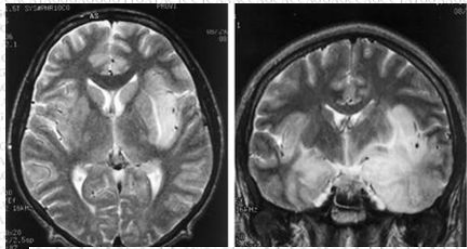
2- HSV antigen detection

- By immunofluorescence or immunoperoxidase staining , or by enzyme immunoassays (EIAs)
- on CSF or detergent-solubilized cells and mucus.

Laboratory Diagnosis of Herpes Simplex Virus

3- Diagnosis of Encephalitis

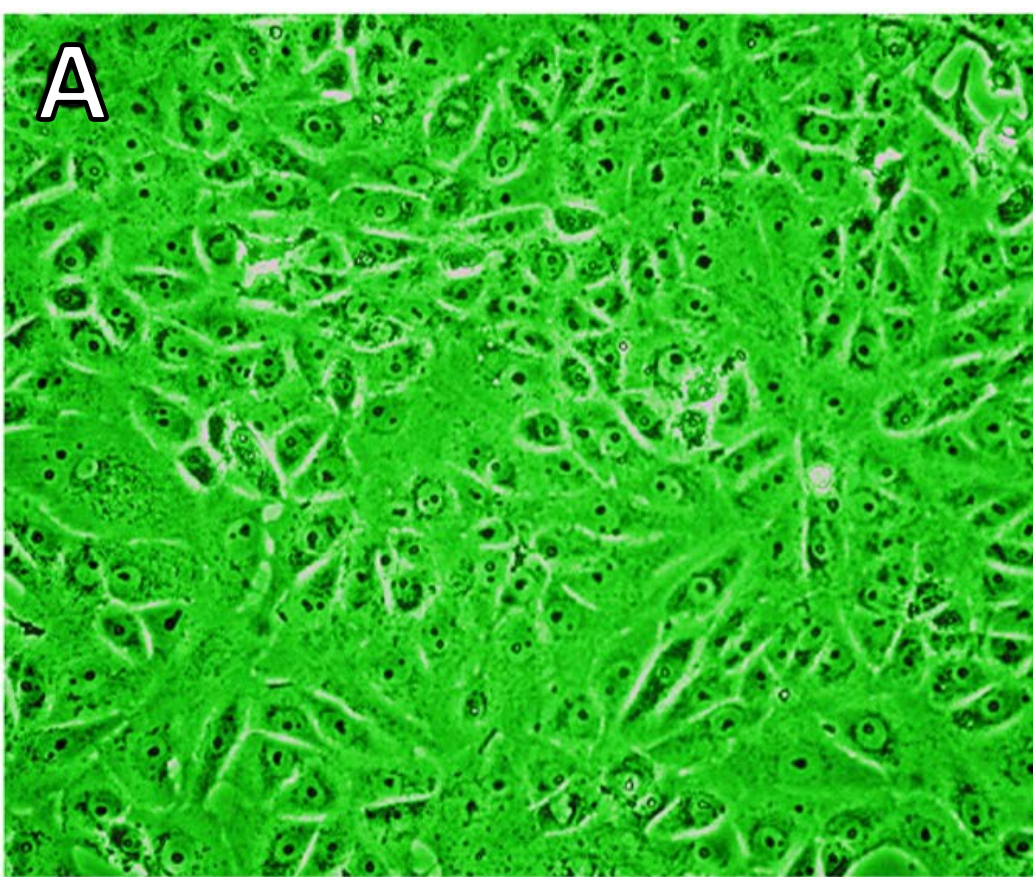
- Previously, brain biopsy
- Currently, PCR is the method of choice for detection of HSV DNA in CSF.



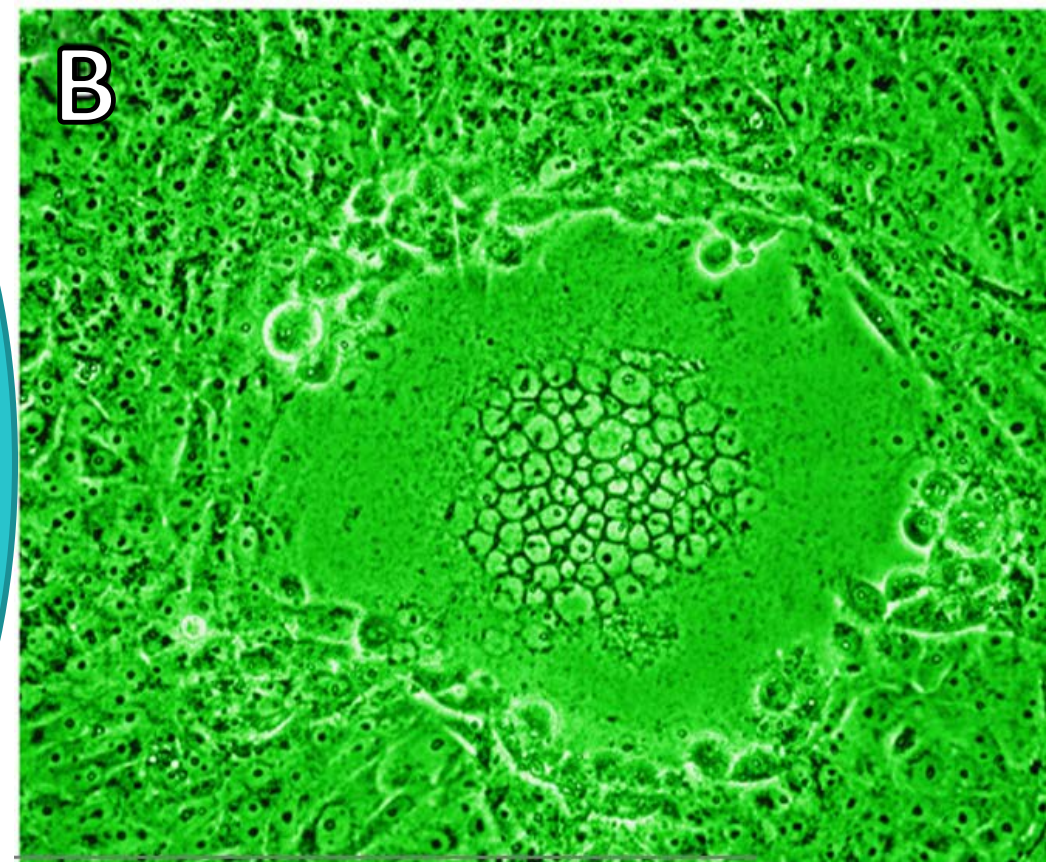
Laboratory Diagnosis of Herpes Simplex Virus

4- Virus isolation in cell culture:

- Currently, used for research purposes, for determining drug resistance where lesions are refractory to treatment
- **Human fibroblasts** or **Vero cells** are both equally sensitive, but HSV replicates rapidly in many mammalian cell lines.
 - **Cytopathic effect (CPE):** Distinctive **foci of swollen, rounded cells** appear within one to five days.
 - The diagnosis can be confirmed within 24 hours by immunofluorescent staining of the infected cell culture.



CPE of
HSV on
Vero
Cell line

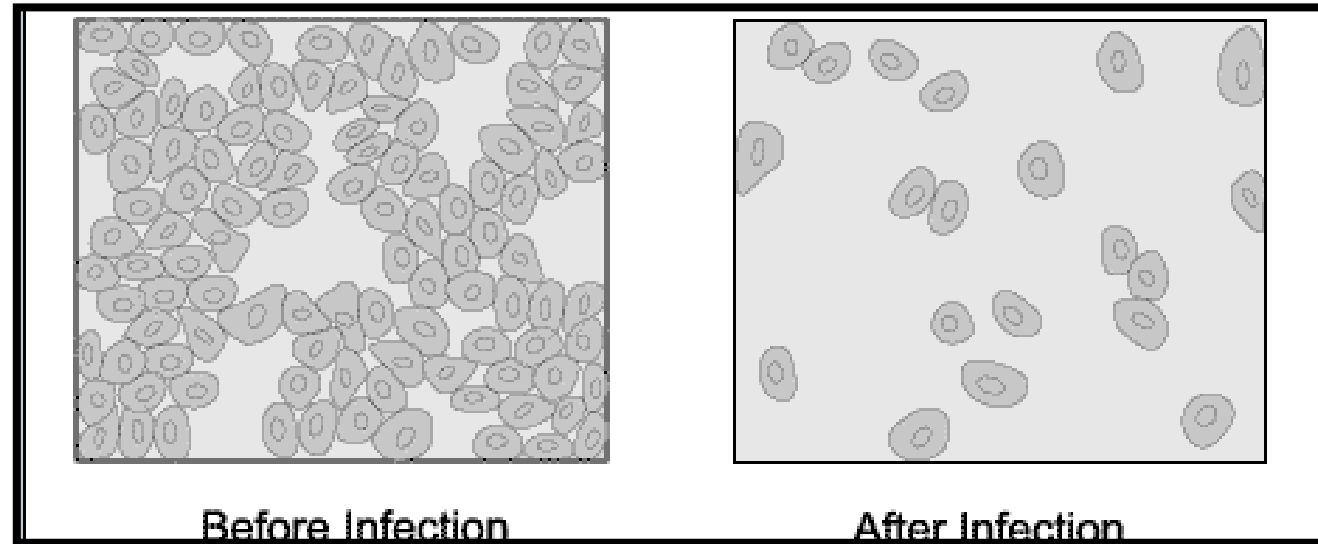


(A) Confluent monolayer of uninfected Vero cells.

(B) Vero cells infected with herpes simplex virus 1 (HSV-1) at 24 hours post-infection. A single large syncytium of at least 50 cells (massed nuclei) which is unstable and later progresses to a necrotic/apoptotic CPE.

Phase contrast microscopy with monochromatic green light provides better resolution.

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1st Assignment: due date 19-2-2018

**Choose two methods for diagnosis of certain human virus
one molecular while the other is serologic method?**