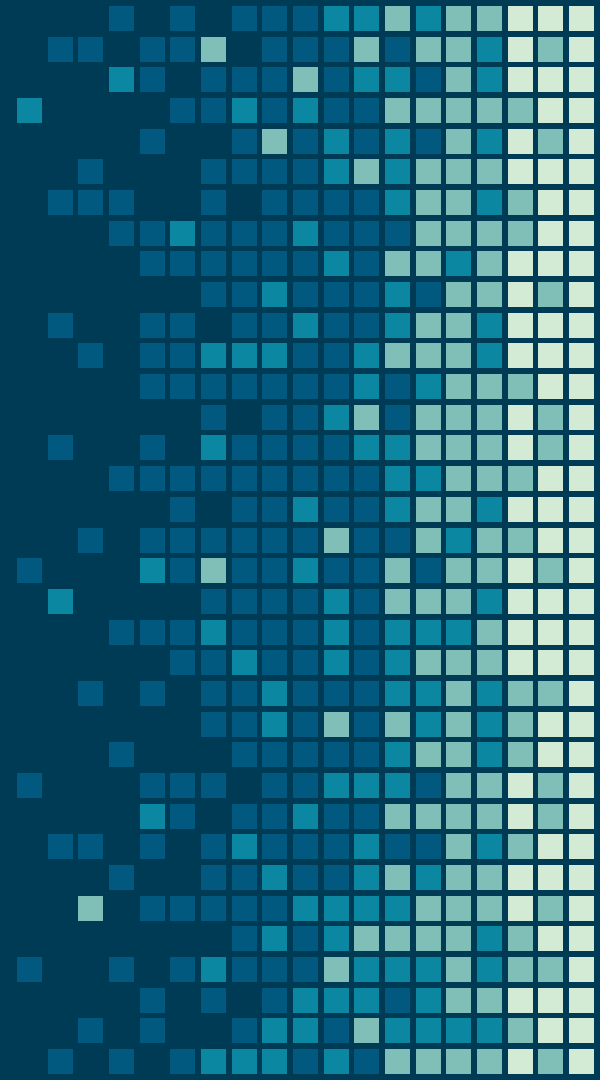
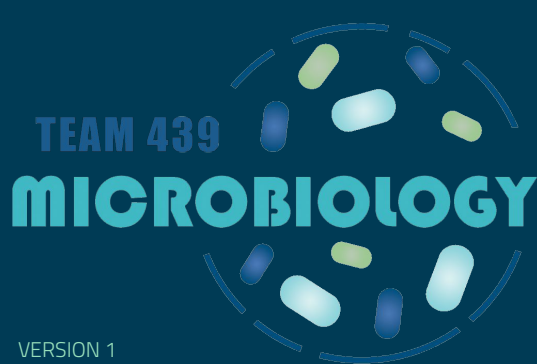


Viral Structure & Classification

(Introduction to viruses)



Objectives

- ❖ Distinguish the viruses from other microorganisms.
- ❖ General characteristics of viruses.
- ❖ Structure & symmetry of viruses.
- ❖ Classification of viruses.
- ❖ Steps of virus replication.
- ❖ Laboratory diagnosis of viral infections.

Colour index:

- **Red: Important.**
- Grey: Extra info & explanation.
- **Purple: only in girl's slides.**
- **Green: Only in boy's slides.**
-

Any future corrections
will be in the editing
file, so please check it
frequently.

Scan the code
Or click [here](#)



Properties of Microorganisms

Characteristic	Parasite	Fungi	Bacteria	Virus
Cell	Yes	Yes	Yes	No
Type of nucleus	Eukaryotic	Eukaryotic	Prokaryotic	-----
Nucleic acid	Both DNA & RNA	Both DNA & RNA	Both DNA & RNA	DNA or RNA
Ribosomes	Present	Present	Present	Absent
Mitochondria	Present	Present	Absent	Absent
Replication	Mitosis	Mitosis Or budding	Binary fission (Bacteria=Binary fission)	Special (one viral particle produces millions of viruses)



Characteristics of Viruses

1

Acellular Organisms

Viruses are not cells.

2

Tiny particles

Can only be seen via E.M

- Size: 20-300 nm
- Unit of viruses: **nm** (10^{-9})
- ~1000 smaller than bacteria)

3

Obligate intracellular organisms

It cannot reproduce outside host cell

No ribosomes or mitochondria, so
¹reproduction, ²protein synthesis, and
³energy production is entirely reliant on host
cell machinery (intracellular resources).

4

Replicate different from cells

One virus can produce millions
of **many viruses**.



Viral Structure

Viral Structure

1- Viral genome

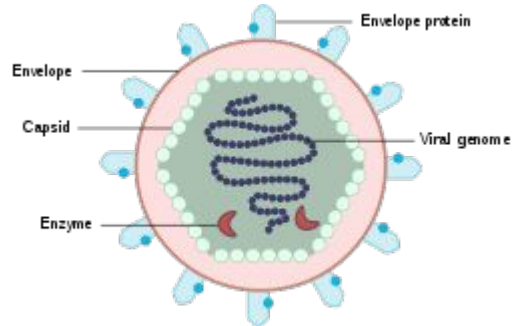
Internal core of viral genome
(containing **DNA or RNA**).

2- Capsid

Protective protein coat that surrounds the nucleic acid.

3- Envelope

Some viruses have lipoprotein membrane.

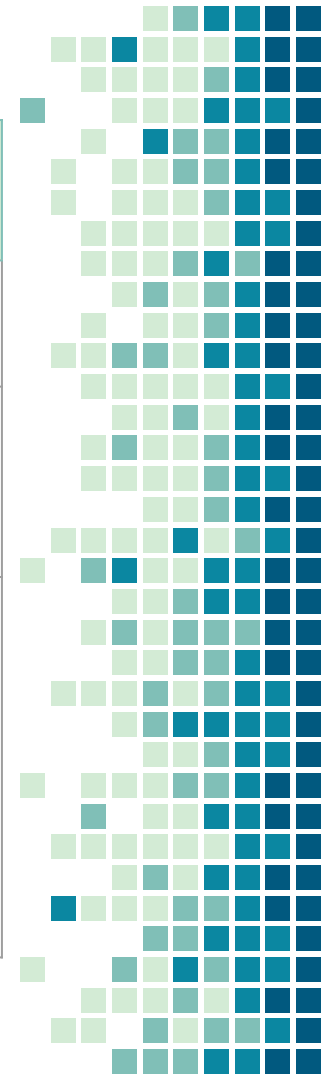


1-Viral Genome (genetic material)

DNA Viruses (Deoxyribonucleic acid)	RNA Viruses (Ribonucleic acid)
All are double stranded <u>Except:</u> Parvoviruses (single stranded)	All are single stranded (ex: Retroviruses) <u>Except:</u> Reoviruses (double stranded)
Single Molecule (Haploid) Meaning: one molecule of double stranded DNA	Number of molecules: <ul style="list-style-type: none">• Single (Haploid)• Double (Diploid ex: Retroviruses)• Multiple pieces of RNA (ex: Reovirus and Influenza)
-	Polarity/Sense: After infection, viral genome: <ul style="list-style-type: none">• (+) polarity/sense acts directly as mRNA (directly translated into protein).• (-) polarity/sense must first be transcribed to mRNA then can be translated into protein.<ul style="list-style-type: none">○ (-)polarity viruses contain virus-specific enzymes for transcription (transcriptase) called: RNA dependent RNA polymerase.

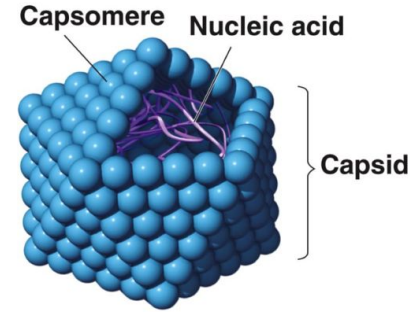
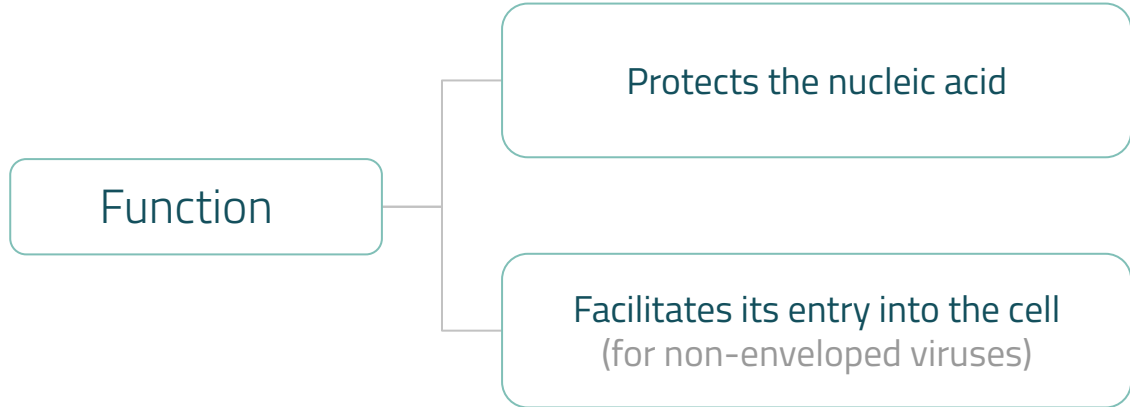


All viruses are **haploid** (have a single copy of genetic material)
Except: **Retroviruses** ex: *HIV* (Diploid: have two copies of the same genetic material)



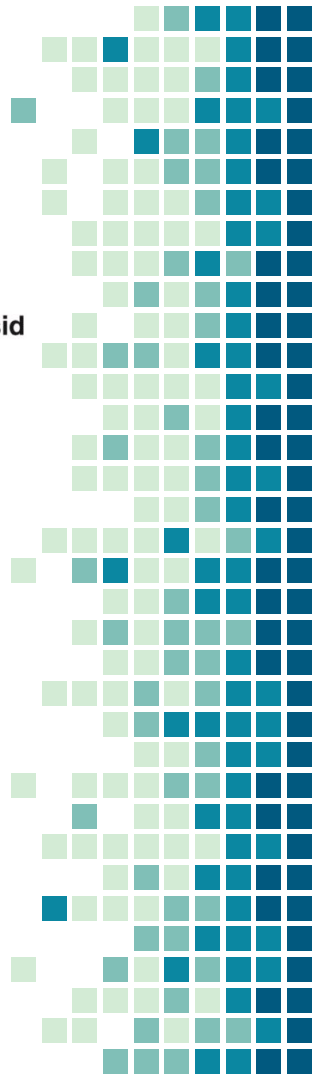
2- Capsid (Protein Coat)

- ❖ A **protein** coat that surrounds the viral genome.
- ❖ Made up of subunits called **capsomeres**.
- ❖ **Nucleocapsid** = nucleic acid (genome) + capsid
- ❖ **Arrangement of capsomers** in capsid gives viruses their **symmetry**.



(a) A polyhedral virus

Note:
All Viruses have a capsid
but not all have a nuclear
envelope

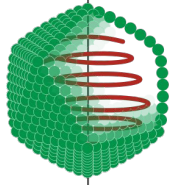


Symmetry:

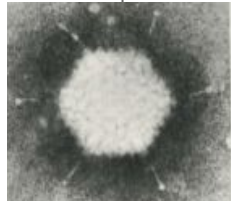
Note: know shapes only (no need to memorize examples).

Based on **arrangement of capsomeres**

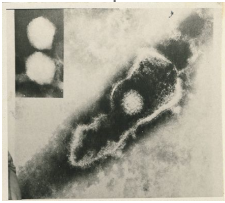
Cubic (Icosahedral)
20 triangles that form an icosahedron, spherical in shape



A. Non-enveloped:
Adenovirus



B. Enveloped:
Herpesvirus



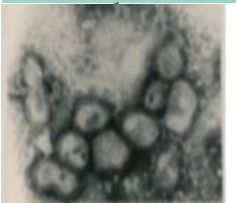
Helical



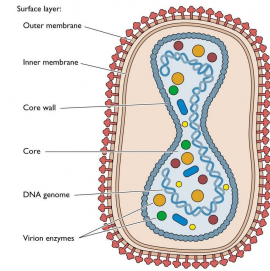
Elongated Filoviruses



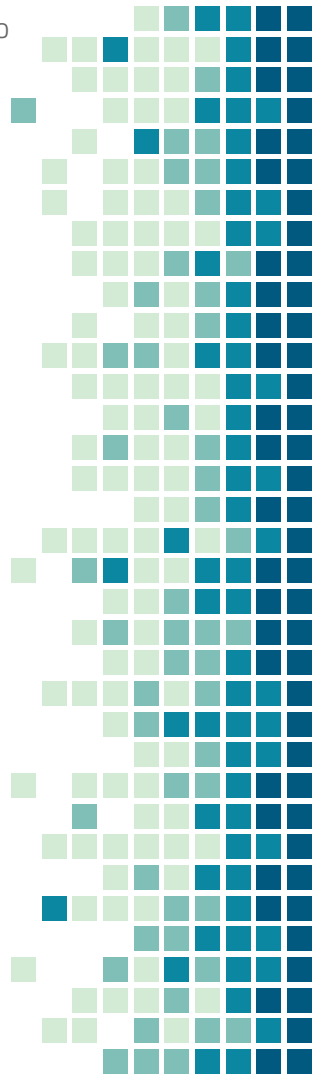
Pleomorphic Influenza v.



Complex

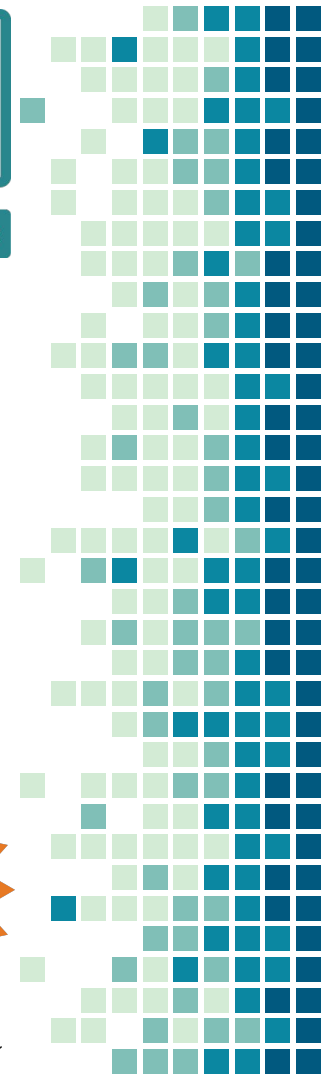
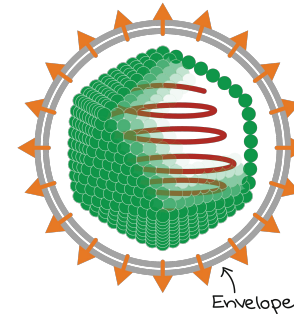
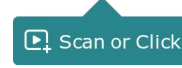


Poxviruses



3- Envelope (Found in some viruses only)

- ❖ **Enveloped viruses are more sensitive** to harsh environmental factors such as : heat and dryness (**Non-enveloped viruses are more resistant**).
- ❖ Envelope is acquired when the virus is released from infected cell (process is called *budding*).
- ❖ **Envelope is derived from host cell membrane**.
 - **Except:** envelope of **Herpes Viruses** which is derived from host **nuclear membrane**.
- ❖ Envelope is a **lipoprotein** membrane
 - **Lipid** from host membrane
 - **Protein/glycoprotein** is virus-specific (from the virus itself)
- ❖ **Glycoproteins:**
 - **attaches to cell receptor** (mediate the attachment of virus to host cell)
 - Virus-specific: specific glycoproteins attach to specific cell receptors
 - Glycoproteins are usually in the form of **spike-like projections**



Viral Proteins

Internal Viral Proteins

➤ Structural proteins:

Capsid proteins (capsomere) are considered internal proteins **in enveloped viruses**.

➤ Non-structural proteins (enzymes):

- **transcriptase** enzyme: *RNA dependent RNA polymerase* (an enzyme that catalyzes the replication of **RNA** from an **RNA** template)

is in all single stranded RNA viruses with negative polarity

- **reverse transcriptase** enzyme (an enzyme used to generate complementary **DNA** from an **RNA** template)

is in Retroviruses and HBV (Hepatitis B Viruses)

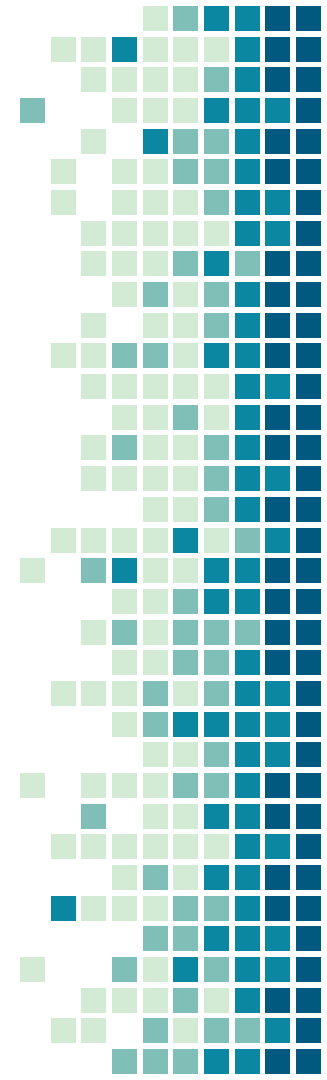
Outer Viral Proteins

➤ **glycoproteins** on envelope:

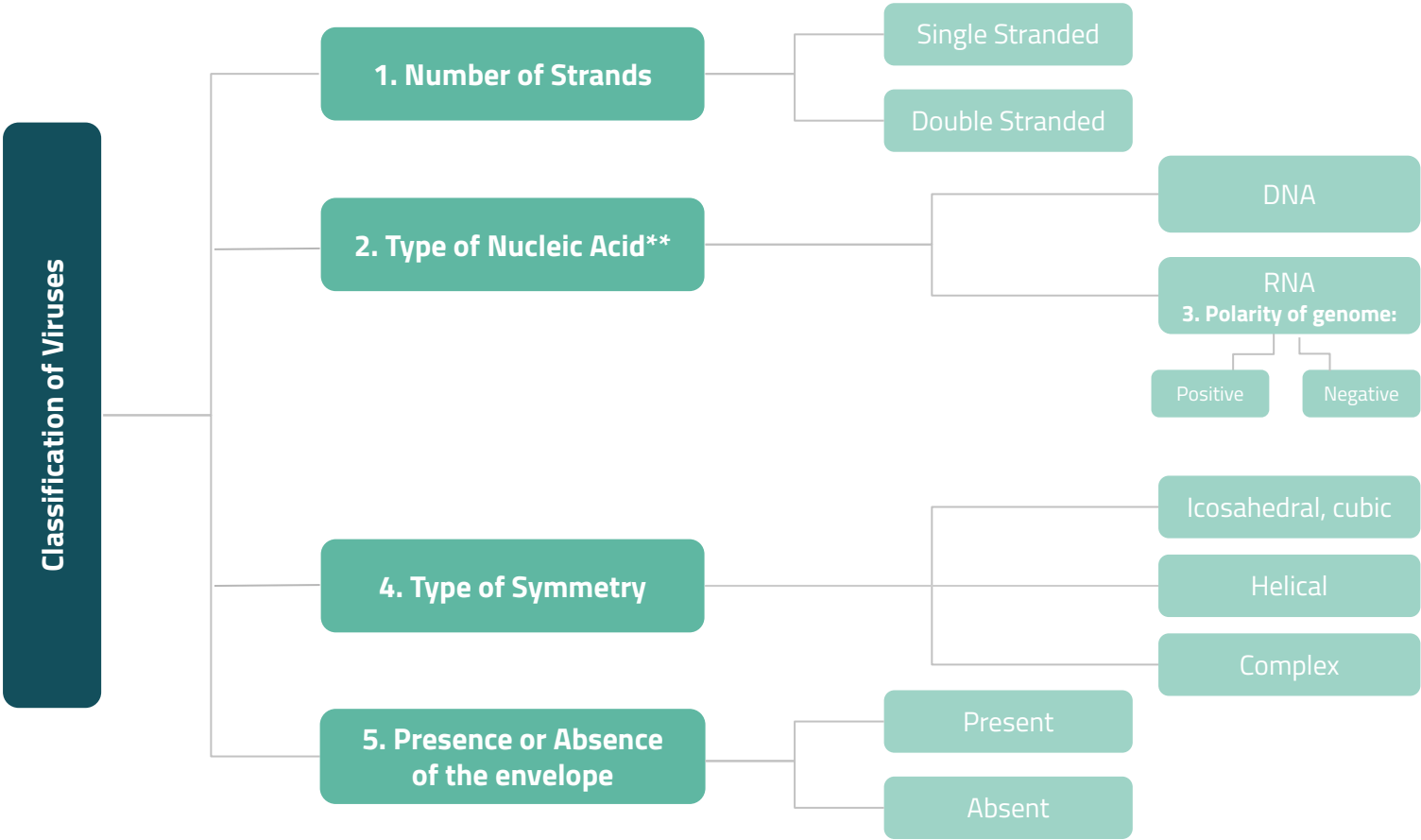
- **Mediate attachment to specific receptors on the host cell.** (specific glycoprotein binds to specific cell receptor)

- **Induce neutralizing antibodies.** (act as antigen; stimulates immune system to produce antibodies.)

- **Are a target of antibodies**



Classification of Viruses



Medically Important Viruses

1-DNA			
Single stranded	Double stranded		
Non-enveloped	Enveloped		Non-enveloped
Icosahedral	Complex	Icosahedral	Icosahedral
Parvoviridae	Poxviridae	Herpesviridae	Adenoviridae

Notice that in DNA viruses, there is no helical symmetry

And that there is no complex symmetry in RNA viruses

2-RNA			
Single stranded			Double stranded
Negative strand	Positive strand		Non-enveloped
Enveloped	Enveloped	Non-enveloped	Icosahedral
Helical Filoviridae	Helical Coronaviridae Icosahedral Retroviridae	Icosahedral Hepeviridae	Reoviridae



Virus Replication Steps (it is recommended to watch [both](#) videos)



Scan or Click

1-Adsorption (Attachment)

Attachment site:

- ❖ **Glycoprotein** (enveloped)
- ❖ **Folding in the capsid proteins** (non-enveloped)

2-Penetration (Entry)

There are two methods:

- ❖ **Fusion** (enveloped). Viral envelope fuses with host cell membrane and enters the cell.
- ❖ **Endocytosis** (enveloped: viruses fuse with endosome membrane),(non-enveloped: virus pores or lyses endosome membrane).

3-Uncoating

- ❖ Removal of capsid
- ❖ Release of viral genome in: **cytoplasm** (for RNA viruses) or **nucleus** (for DNA viruses).

4-Synthesis of Viral components (mRNA, viral proteins, N.A.)

- ❖ **Positive Single Stranded RNA** viruses, their single strand of RNA acts directly as mRNA (**Direct**) which will be translated (with help of the cell's ribosomes) to proteins & enzymes.
(NO NEED for transcription, positive single stranded RNA is translated directly to make viral proteins)
- ❖ **Negative RNA** viruses, on the other hand, their RNA must be **transcribed** into mRNA first by **RNA dependent RNA polymerase** which is then translated to make viral proteins (**Indirect**).
- ❖ Viral genome is replicated in both cases.

5-Assembly

- ❖ **(Nucleic acid + Viral proteins= Virions)**
Nucleic acid and viral proteins assemble to make **virions** (an entire viral particle).
Millions of virions are produced.

6-Release

- ❖ **Enveloped viruses:** undergo **budding** and get their membrane either from the:
¹**cell membrane** (in RNA viruses) or ²**nuclear membrane** (in DNA viruses e.g herpes virus).
- ❖ **Non-enveloped viruses:** virus **lysis** or ruptures from the cell membrane (**cell is damaged**).

Microscopic Examination

Cell Culture

Isolation of virus and diagnosis

Laboratory Diagnosis of Viral Infections

Viral Antigen/Antibody Detection

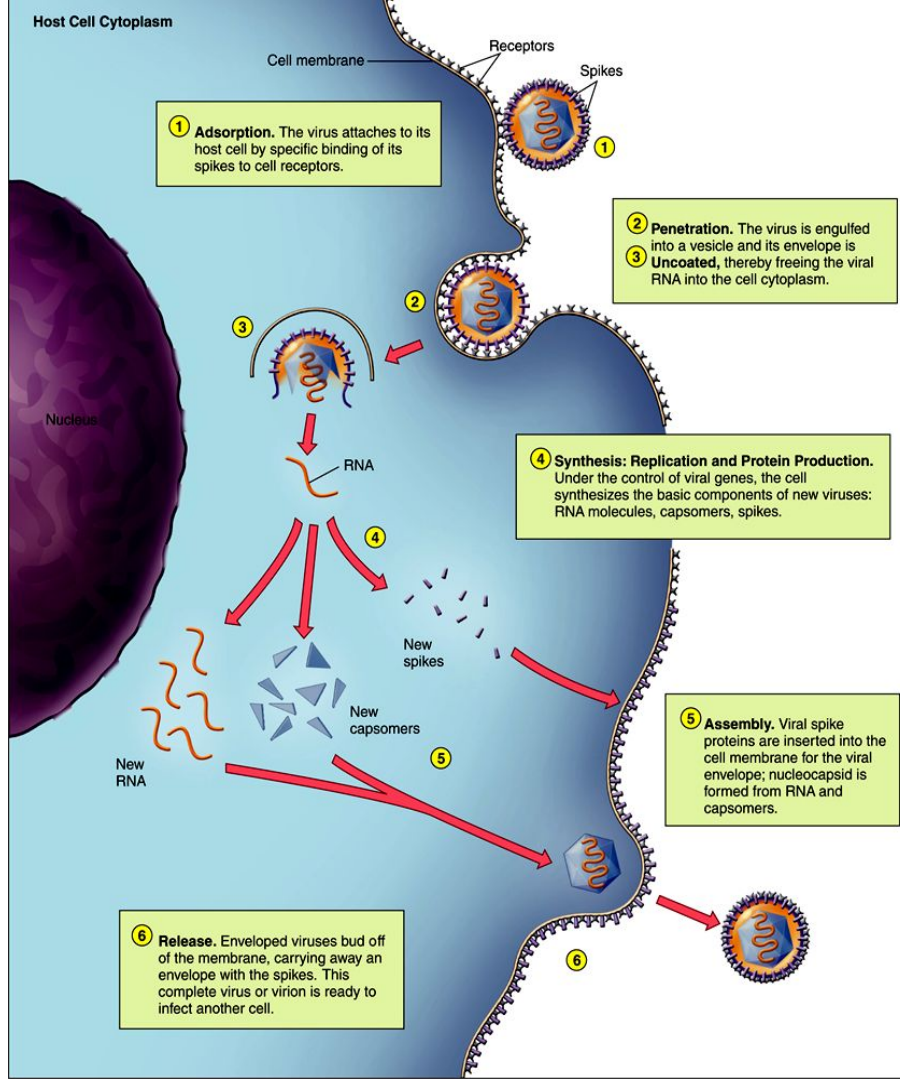
Routinely used

Molecular Method

Most expensive, not routinely used

Serological Tests

Routinely used, for diagnosis

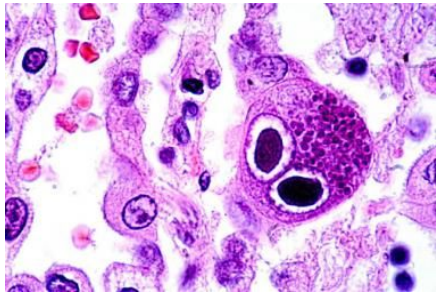


Microscopic Examination

Light Microscopy

- Shows **histological appearance** and **cytopathic effect**
(to see the effect of the virus on the cells **not the virus itself**)

E.g. **inclusion bodies.**



Owl's eye (CMV)

Electron Microscopy

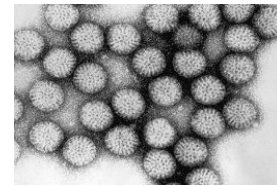
- Shows **size** and **morphology** of the virus
(**to see virus itself**).

- Expensive, so used in research and new viruses

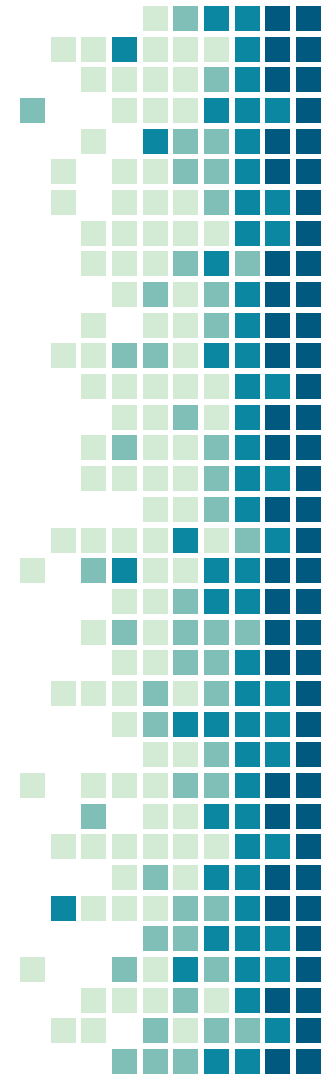
E.g.

- Diagnosis of viral GE (gastroenteritis) such as rota & adenovirus.
- Diagnosis of skin lesions caused by herpes or poxvirus.

- It is replaced by antigen detection & molecular tests.



Rotavirus



Virus cultivation

❖ Methods of Virus Cultivation

1. **Laboratory animals** (effectiveness of vaccine)
2. **Embryonated eggs** (to develop vaccine)
3. **Cell culture** (isolation of virus)

1st step: Cell culture

-Tells whether or not there is infection through **cytopathic effect**. It is not a definitive diagnosis.

2nd step: further investigation to identify pathogen through other tests like:

- Immunofluorescence
- ELISA
- Molecular testing

Cell Culture:

Purpose: isolation of virus (DNA or RNA), new virus diagnosis.
Used to see: cytopathic effect (not virus or type of virus)

Primary cell culture

1 or 2 subpassages
(lasts a very short period which is why it is **difficult to maintain**)

Diploid cell culture (semi-continuous)

20 to 50 subpassages
(lasts for a few weeks)

Continuous cell line

Indefinite
(**cancerous cell line**)

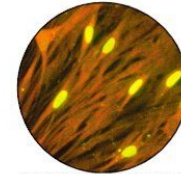
Problems regarding cell culture:

- **Long incubation** (5 days or more), it can be solved with rapid culture technique.
- **Sensitivity is variable.**
- Susceptible to **bacterial contamination.**
- Some viruses **don't grow in cell culture** (HCV -Hepatitis C virus)



Rapid cell culture technique:

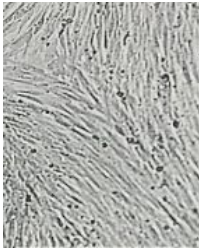
- A modification of cell culture called **Shell vial assay**
- Based on immunofluorescent **detection of viral antigens**
- 1-3 days



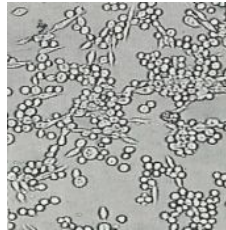
Immunofluorescence

Detection of Viral Growth

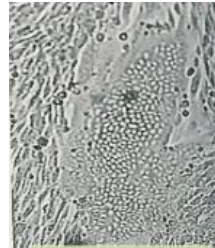
1. **Cytopathic effects:** the affected cell will appear round, shrink and aggregated (in a group). Syncytium (giant multinucleated cell) and loses adherence. **Basis of cell culture.**



Uninfected CC



Cell rounding



Syncytium

2. **Immunofluorescence (IF)** basis of Shell vial assay
3. Other methods.

Serological Tests

-Used specifically for viruses to detect:

- Antigen
- Antibody

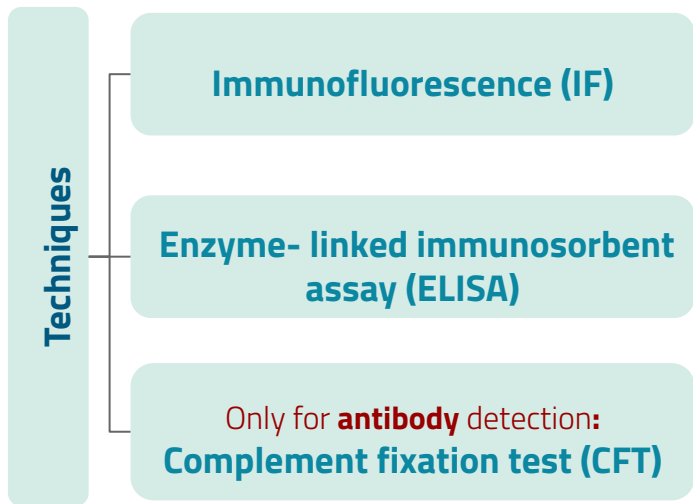
-Examples of Serological tests:

- Immunofluorescence
- ELISA

Antibody and Antigen Detection

(**Indirect** indication of viral infection)

(**Direct** indication of viral infection)



Antigen detection

It is a test that looks for (**antibodies**) from a sample to determine the availability of (**antigen**).

Test	Sample	Virus
IF	Nasopharyngeal aspirate	Influenza V
	Skin scrapings	HSV
ELISA	Faeces	Rotavirus
	Blood	HBV(HBsAg)

Immunofluorescence

ELISA

(Enzyme-Linked Immunosorbent Assay)

Molecular Test

Uses **fluorescent microscope** and antibodies labeled with **fluorescent dye**

Look through a fluorescent microscope:

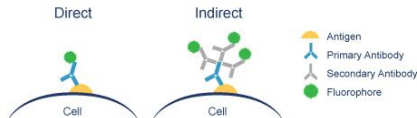
- If green fluorescent: positive result
- No fluorescence: negative result

❖ **Direct IF: antigen** detection

Add antibody labeled with fluorescent dye to cells on slide.

❖ **Indirect IF: antibody** detection

Add secondary-antibodies labeled with fluorescent dye to patient serum.

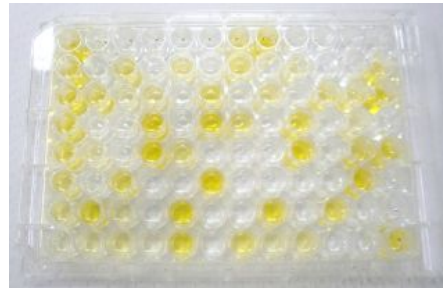


Same principle as IF.

Uses **spectrometer** and antibodies labeled with **enzymes** instead of fluorescent dye.

- Yellow: positive result
- Colorless: negative result

- **Direct: antigen** detection.
- **Indirect: antibody** detection.



The coloured cells indicate reactivity

Uses **Polymerase Chain Reaction**

- Amplifies viral genome and nucleic acid
- **Used for diagnosis** as a confirmatory test.
- It is **the only way to monitor a patient's response to treatment** by measuring viral load/concentration.

MCQs

1- Viral symmetry is determined by:

- a) Viral envelope
- b) Viral Capsid
- c) Viral genome
- d) Viral Enzyme

2-Which of the following is true about viruses?

- a) They have either DNA or RNA
- b) They reproduce by binary fission
- c) All of them have an envelope
- d) They are made up of one cell

3-All viruses are haploid except:

- a) Influenza
- b) Parvoviruses
- c) Reoviruses
- d) Retroviruses

4-Which of the following is directly translated into protein inside the host cell?

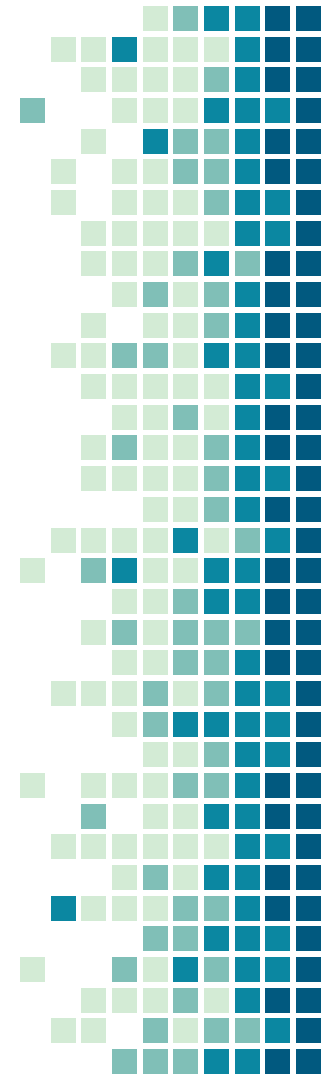
- a) Positive ddDNA
- b) Negative ssDNA
- c) Positive ssRNA
- d) Negative ddDNA

5-Which of the following is not an internal viral protein?

- a) Transcriptase
- b) Reverse transcriptase
- c) Capsid proteins in enveloped viruses
- d) Glycoproteins

6-Viruses are considered obligate parasites because they?

- a) Are acellular
- b) Are tiny particles
- c) Need a host to complete their life cycle
- d) None of the above



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MCQs

7-To see the virus which method is used?

- a) Light microscope
- b) Cell Culture
- c) Electron microscope
- d) Fluorescence

8-Which if the following is a virus cultivation technique used to isolate the virus?

- a) ELIZA
- b) Embryonated eggs
- c) Cell cultures
- d) Laboratory animals
- e) Polymerase Chain Reaction

9-Which is not an example of a serological test?

- a) Complement Fixation Test
- b) Immunofluorescence
- c) ELIZA
- d) FISH
- e)None of the above

10-Which of the following is used to monitor a patient's response to treatment by measuring viral load?

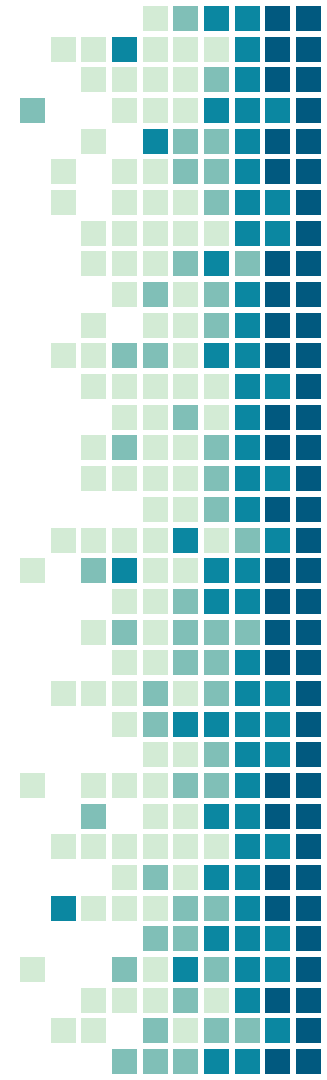
- a) Complement Fixation Test
- b) Immunofluorescence
- c) Polymerase Chain Reaction
- d) ELIZA
- e)None of the above

11-which of the following is not a structural classification of viruses?

- a) complex
- b) vibro
- c) helical
- d) isocahedral

12- The presence of which of the following is a direct indication of a viral infection:

- a) Antibody
- b) Antigen
- c) ribosomes
- d) endotoxins
- e) chemotaxins



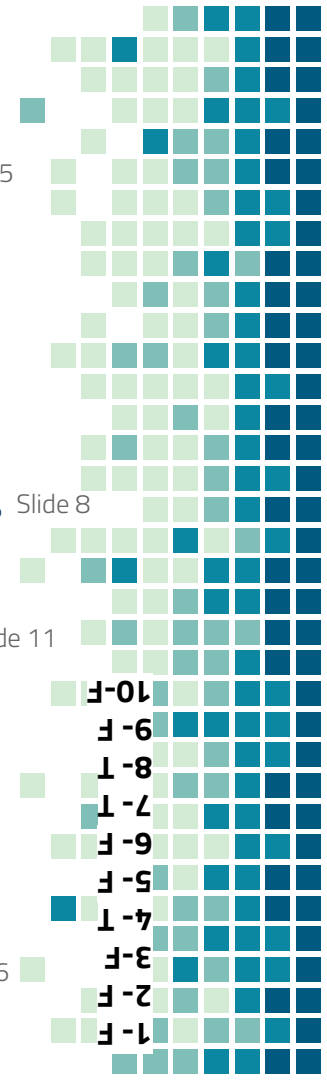
q - 21
q - 11
c - 01
p - 6
c - 8
c - 7

T & F

1. All viruses can be grown in cell cultures. ()
2. Complement fixation test is used for antigen detection. ()
3. Immunofluorescence uses antibodies labelled with enzymes. ()
4. Molecular tests are used to amplify viral genome and nucleic acids. ()
5. Primary cell cultures can last for several weeks. ()
6. Electron Microscopy shows cytopathic effect and histological appearance. ()
7. Non-enveloped viruses leave the host cell by lysing it. ()
8. Viral replication produces millions of viruses from one virus. ()
9. Transcriptase enzyme is found in retroviruses. ()
10. Capsids are derived from host membrane. ()

SAQ

- 1- List the three main viral structure components. Slide 5
- 2- What is a nucleocapsid? Slide 7
- 3- What are the functions of viral capsids? Slide 7
- 4- What are the types of symmetry that viruses exhibit? Slide 8
- 5- What are the 5 criteria for virus classification, and which criteria is only present in RNA viruses? Slide 11
- 6- What is an example of viruses that contain an envelope that is derived from the host's nuclear membrane? Slide 9
- 7- What are the problems regarding cell culture? Slide 16



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