

Microbiology – Lecture 7

Introduction to viruses



TEAM 437

Red: important

Green : doctor notes

Black : original slides

Grey: extra information

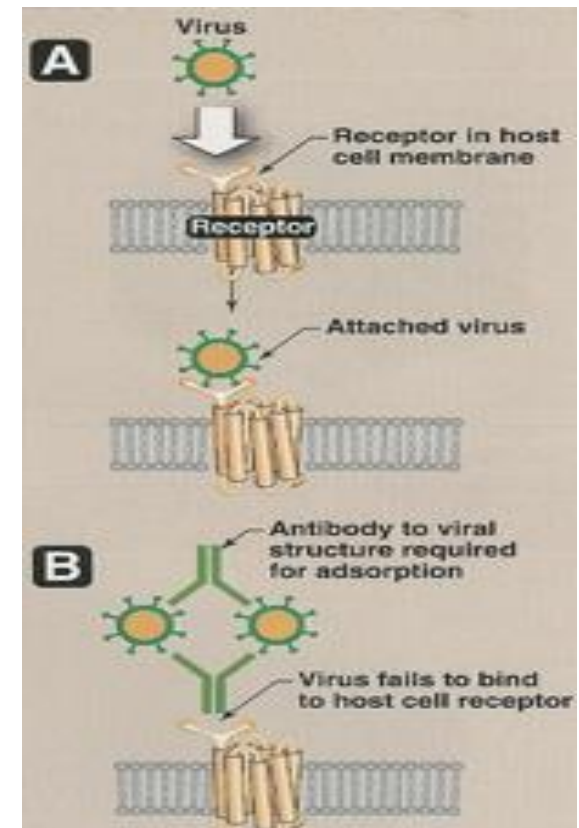
In this link, you will find any corrections or notes unmentioned in the team's work. Please check the link below **frequently**.

https://docs.google.com/presentation/d/1yIQt3G8UDFG6xYMRhXkTk-dS54NeTfhJaPe_y0M-kjk/edit?usp=sharing




Objectives

1. Distinguish the viruses from other microorganisms
2. General characteristics of viruses.
3. Structure & symmetry of viruses.
4. Classification of viruses.
5. Steps of virus replication .
6. Laboratory diagnosis of viral infections.





Properties of Microorganisms

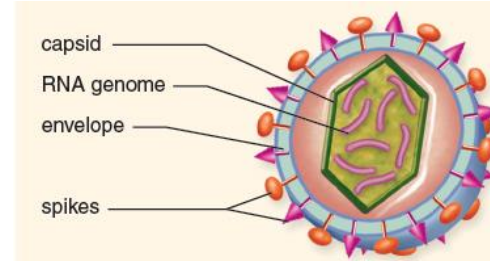
Characteristics	Parasites	Fungi	Bacteria	Viruses
Is it a cell?	Yes	Yes	Yes	No
Type of nucleus	Eukaryotic	Eukaryotic	Prokaryotic	
Type of nucleic acid	Both DNA & RNA	Both DNA & RNA	Both DNA & RNA	DNA <u>OR</u> RNA (not both!)
Ribosomes	Present	Present	Present	Absent
Mitochondria	Present	Present	Absent	Absent
Replication	Mitosis	Budding or Mitosis	Binary fission	Special

Characteristics of Viruses & Virus proteins



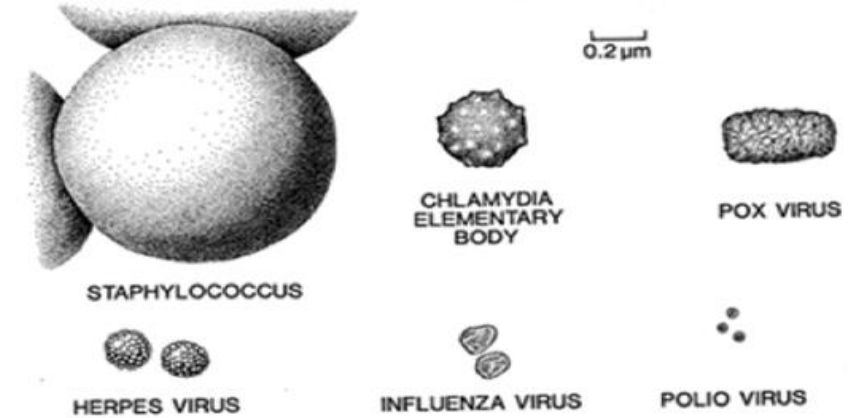
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- They are **Acellular** organisms (Acellular = not having cells)
- They are tiny particles consisting of :
 1. An internal core (Containing DNA or RNA)
 2. Protein coat (capsid)
 3. some viruses also have **lipoprotein membranes (envelope)** the **envelop** is taken from the host .



- They are **obligate intracellular organisms**. (which means that they are unable to reproduce outside of their host cell) **Why? They don't have ribosomes or mitochondria**
- Replicate in a manner different from cells (e.g. one virus replicate to Produce many viruses)
- Size: 20-300 nm

They can only be seen by electron because of their tiny size
They are 1000 times smaller than bacteriamicroscope



The Outer Viral Proteins	The Internal Viral Proteins
<ul style="list-style-type: none"> • Mediate attachment to specific receptors on the host cell • Induce neutralizing antibodies. <small>(neutralizing antibody: is an antibody that defends a cell from an antigen or infectious body by neutralizing any effect it has biologically).</small> • They are the target of antibodies 	<ul style="list-style-type: none"> • Structural proteins (capsid proteins of enveloped viruses) <ul style="list-style-type: none"> • Nonstructural proteins (enzymes) ➤ All single stranded RNA viruses with (-) polarity have transcriptase (an RNA dependent RNA polymerase) inside virions. <small>(Virions: the complete, infective form of a virus outside a host cell)</small> ➤ Retroviruses & HBV contain reverse transcriptase

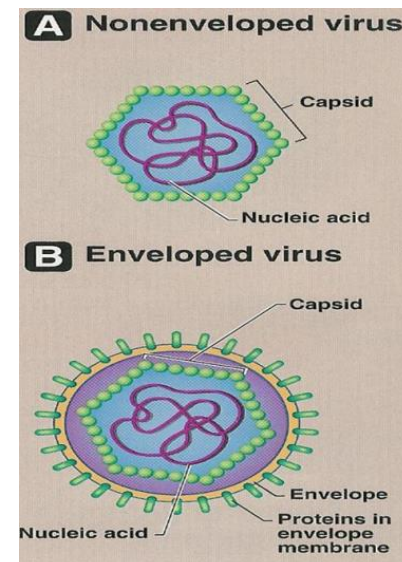
All viruses are haploid,
except Retroviruses which
are diploid

Viral Structure



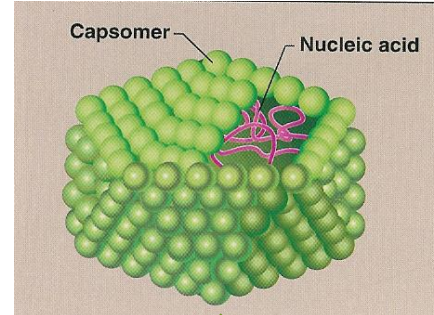
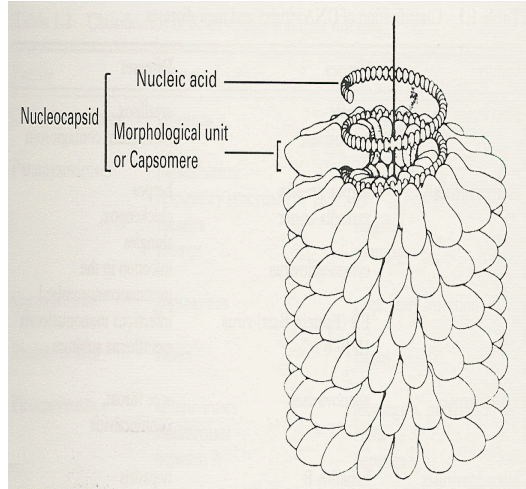
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<u>1-Viral Genome</u> Can be either	<u>2-Envelope</u>	<u>3-Capsid</u>
<p>1-RNA (Ribonucleic acid) All RNA viruses have single stranded RNA <u>except Reoviruses(multiple)</u></p> <ul style="list-style-type: none"> (+) polarity (having same sequence as RNA) (-) polarity (having complementary sequence as RNA) <p>2-DNA (Deoxyribonucleic acid)</p> <ul style="list-style-type: none"> All DNA viruses have double stranded DNA <u>except Parvoviruses</u>. Has single stranded Single molecule (It mean they only have 1 molecule of DNA) 	<p>It is a <i>Lipoprotein membrane</i> (host lipid + virus specific protein) contains protein and glycoprotein</p> <p>Budding</p> <ul style="list-style-type: none"> Envelope is derived from <u>host cell membrane</u>, except herpesviruses from nuclear membranes. Enveloped Viruses are more sensitive to heat ,dry & other factors than non-enveloped Viruses. Glycoprotein attaches to host cell receptor usually in form of spike / finger like projections. 	<ul style="list-style-type: none"> A protein coat made of subunits (capsomeres) Genome (Nucleic acid)+capsid = nucleocapsid <p><u>Function</u></p> <ul style="list-style-type: none"> -Protects Nucleic Acid -Facilitates its entry into cell <p>For more info click here</p> <p>Also here</p>

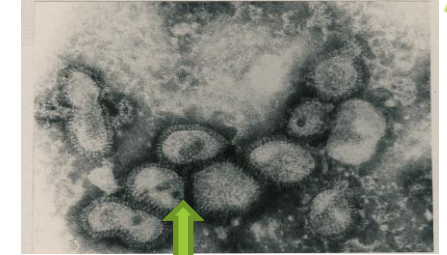




Symmetry



Elongated (filovirus)



pleomorphic (influenzavirus)

helical

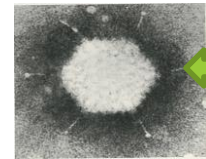
symmetry

complex

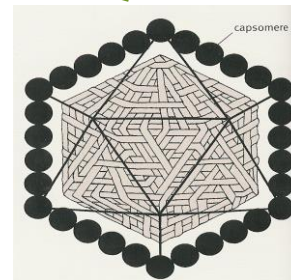
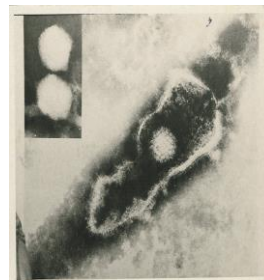
20 Triangles

cubic (Icosahedral)

Adenovirus



Herpesvirus



poxivirus



The arrangement of a virus' capsomeres gives it a unique symmetry



Classification of Viruses

	ENVELOPED	NON-ENVELOPED		
DNA	<p>dsDNA</p> <p><i>Poxviridae</i></p> <p><i>Herpesviridae</i></p> <p><i>Hepadnaviridae</i></p>	<p>dsDNA</p> <p><i>Adenoviridae</i></p> <p><i>Papovaviridae</i></p>		
RNA	<p>ss RNA</p> <p><i>Coronaviridae</i></p> <p><i>Paramyxoviridae</i></p> <p><i>Bunyaviridae</i></p> <p><i>Orthomyxoviridae</i></p> <p><i>Arenaviridae</i></p> <p><i>Togaviridae</i></p> <p><i>Flaviviridae</i></p> <p><i>Retroviridae</i></p> <p><i>Rhabdoviridae</i></p> <p><i>Filoviridae</i></p> <p>100nm</p>	<p>dsRNA</p> <p><i>Reoviridae</i></p>		<p>ssRNA</p> <p><i>Picornaviridae</i></p> <p><i>Caliciviridae</i></p>
	<p>ssRNA</p> <p><i>Picornaviridae</i></p> <p><i>Caliciviridae</i></p>			

Classification of Viruses

Type of Nucleic Acid
(DNA/RNA)

The number of strand
(Single/Double)

The polarity
of viral genome
(+/-)

The presence or
absence of envelope

Type of symmetry



بس اعرفوا التقسيم
No need to
memorize names

Medically Important Viruses

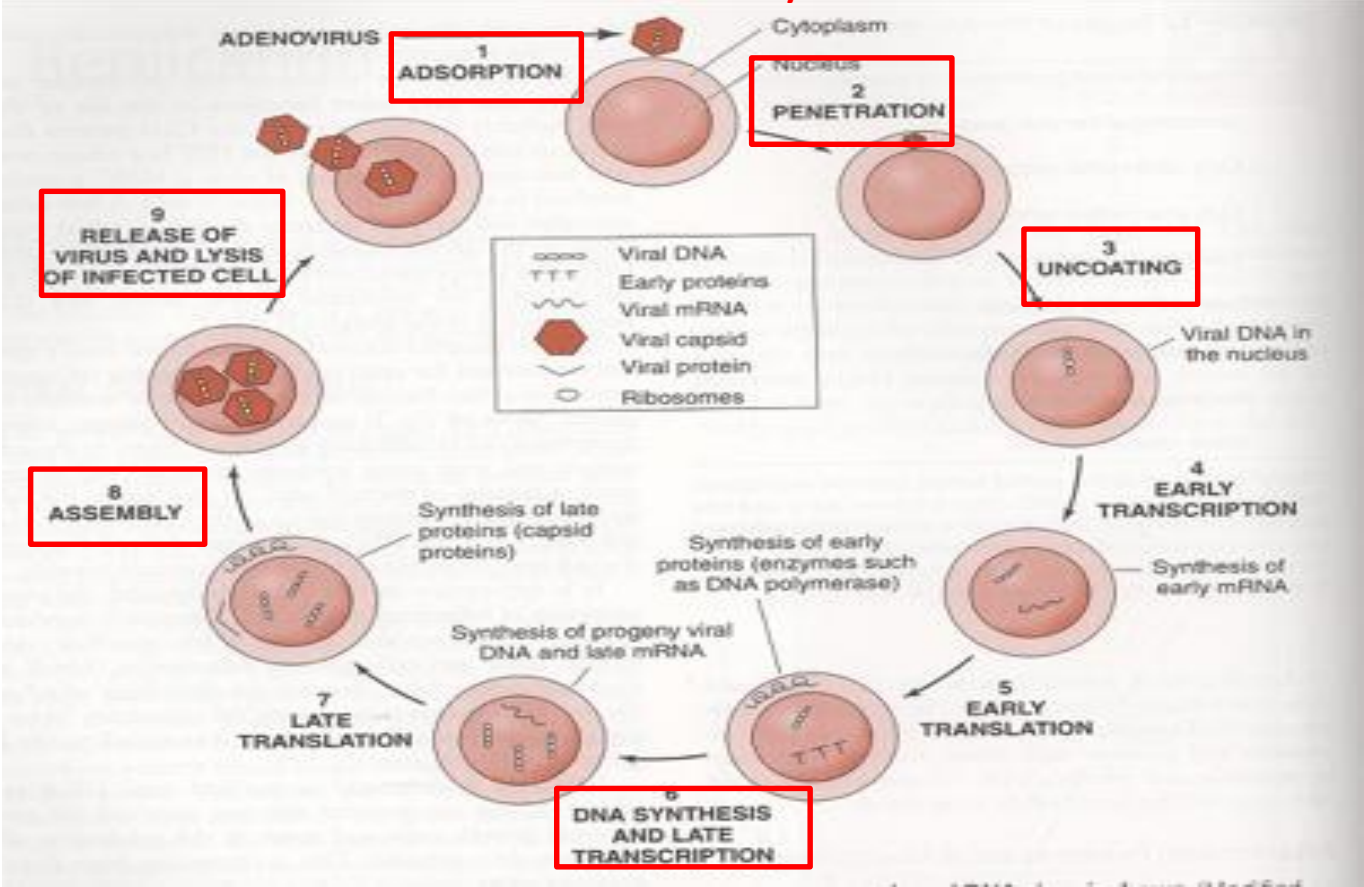
1- DNA			
Single Stranded	Double Stranded		
Non- Enveloped	Enveloped		Non- Enveloped
<u>Icosahedral</u>	<u>Complex</u>	<u>Icosahedral</u>	<u>Icosahedral</u>
Parvoviridae	Poxviridae	Herpesviridae	Adenoviridae

2- RNA			
Single Stranded			Double Stranded
Neg-Strand	Pos-Strand		Non-Enveloped
Enveloped	Enveloped	Non-Enveloped	<u>Icosahedral</u>
<u>Helical</u> Filoviridae	<u>Helical</u> Coronaviridae	<u>Icosahedral</u> Hepeviridae	Reoviridae
	<u>Icosahedral</u> Retroviridae		



Replication

Viral Growth Cycle



1-Adsorption
(Attachment)

6-Release

2-Penetration

5-Assembly

3-Uncoating

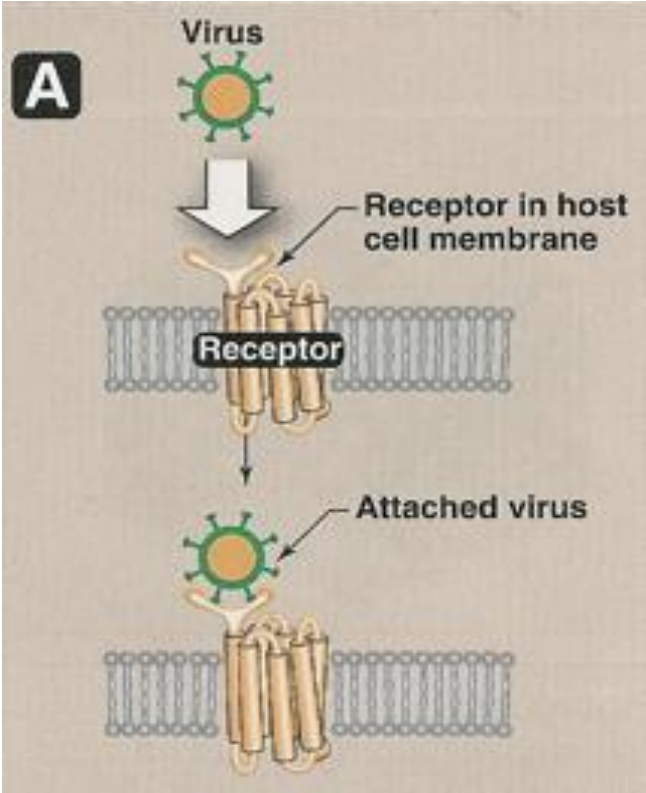
4-Synthesis of viral
components
(mRNA,Viral proteins,
Nucleic Acid)



Replication

1-Adsorption

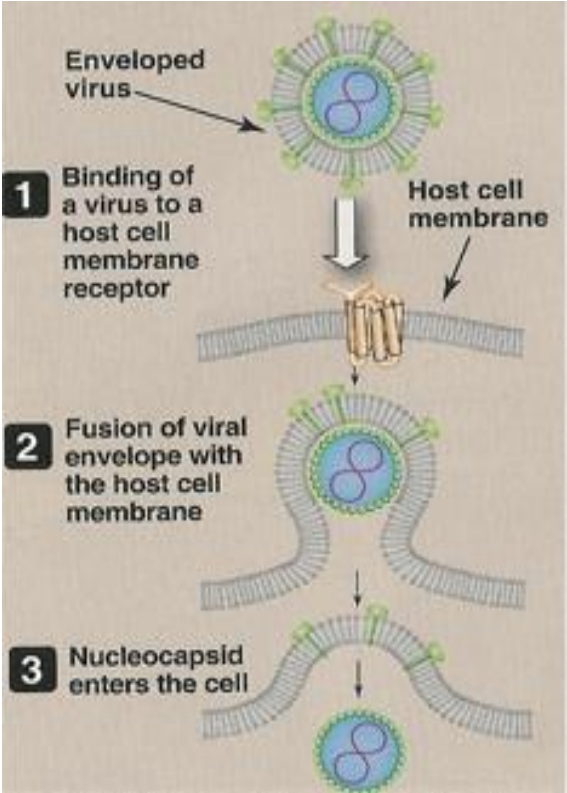
ex- glycoprotein fiber



2-Penetration

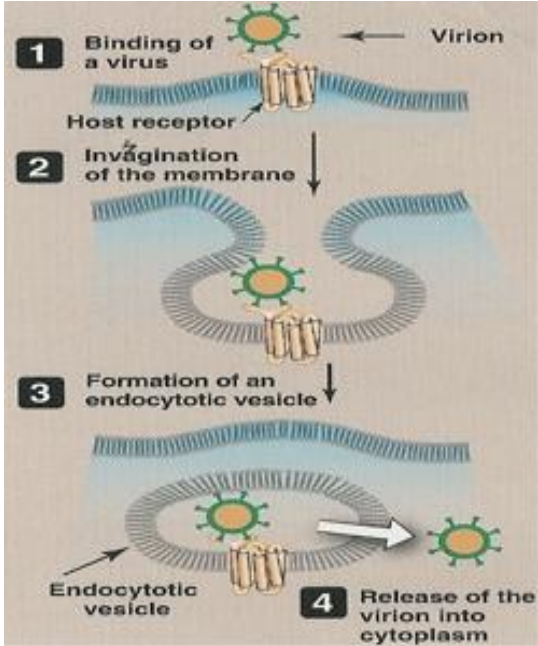


1-Fusion
(for enveloped virus only)



2-Endocytosis (cell eating)

- A. Enveloped virus fuses with endosome membrane
- B. Nonenveloped Virus lysis, pore



Note: Why can the enveloped virus membrane infuse with cell membrane?

It is made of lipids unlike bacteria made of polysaccharides

Replication



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3-Uncoating

Uncoating:

- Release of viral genome
- cytoplasm
- nucleus (Most DNA viruses replicate here)

5-Assembly

The viral Proteins +Nucleic acid= **virion**

Then the virion is released.

What is a **virion**? is a complete virus particles, consisting of RNA or DNA surrounded by a protein shell.

Basically it is the infective form of a virus.

4-Synthesis of viral components

+ssRNA = single stranded +RNA

1. mRNA

Viral genome $\xrightarrow{\text{transcription}}$ mRNA
 $\xrightarrow{\text{+ssRNA acts directly}}$

mRNA

2. Viral proteins

mRNA $\xrightarrow{\text{translation}}$ viral proteins:
cell ribosome

- enzymes
- structural proteins
(eg; capsomeres)

3. replication of viral genome

- The virus will replicate and synthesize it's nucleic acids using the host cell's resources مواردھا
- One virus can produce millions of viruses by continuation of this process (synthesis of viral proteins)

If –RNA virus : viral genome goes through transcription to make (+mRNA) which is translatable. So it goes through ALL the steps

If +RNA virus : no need for step 1 (already has mRNA) so it automatically goes to step 2.

Replication



last step of the replication cycle of the viruses.

6- Release

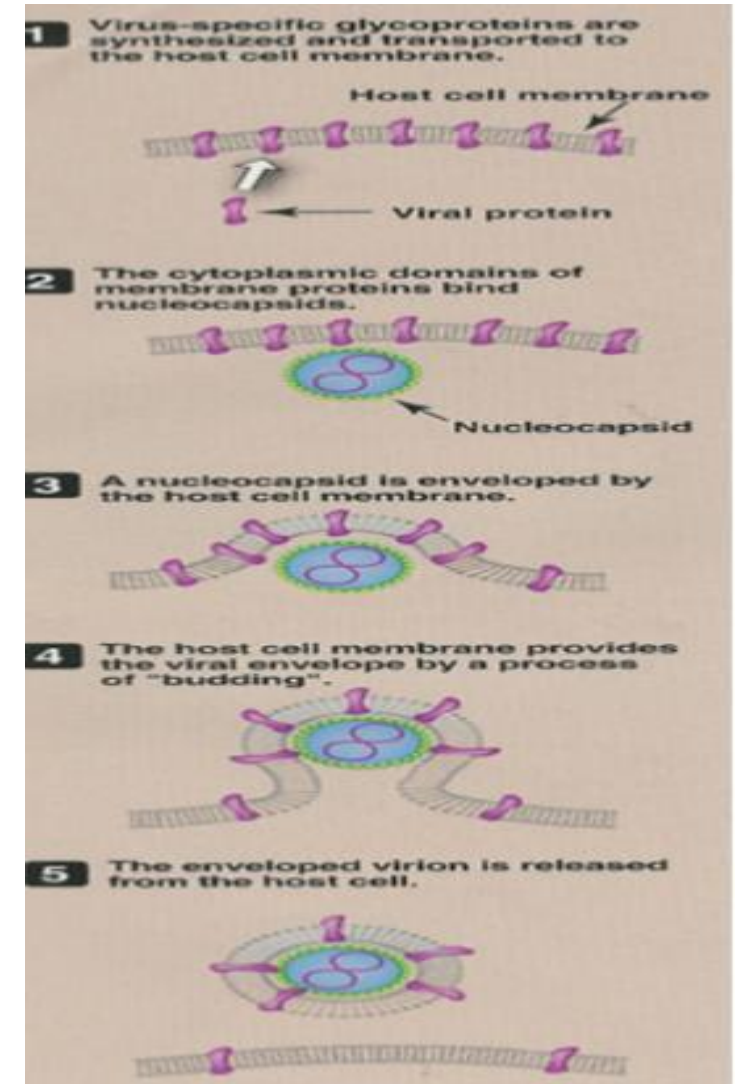
Enveloped Viruses

Non-Enveloped viruses

The Virion leaves the Cell and creates its Envelope by **budding** through the host's cell membrane.

Except **Herpes** viruses = budding host's nuclear membrane

Cell **lysis** or **rupture**





Laboratory Diagnosis of Viral Infections

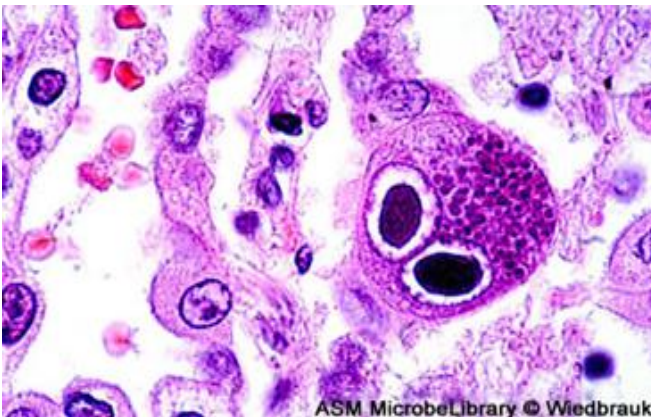
Microscopic examination.

❖ Light microscopy:

See effect of virus on cell
Can't see Virus

Histological appearance "to see
what the virus has done to the cell,
without seeing the virus itself"

Ex. "Inclusion bodies"



Owl's eye (CMV)

❖ Electron microscopy:

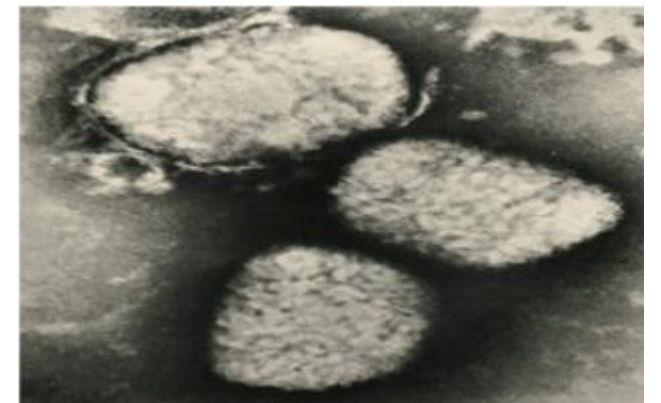
see **SIZE** and **Morphology** of virus.

Morphology & size of virions . " to see the virus
particle itself"

- Ex. Diagnosis of viral gastroenteritis such as Rota, adenoviruses. Diagnosis of skin lesion caused by herpes, or poxviruses.
- It is replaced by Antigen detection & molecular tests



Herpesvirus



Poxvirus

Difficult to handle used only in laboratories

Virus cultivation :

- a) Laboratory animal.
- b) Embryonated egg.
- c) Cell culture.

Cell culture



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- Cell culture refers to the **removal of cells** to see their subsequent **growth** under suitable environment.
- **After isolating** the cells from the tissue, it has 3 types of sub passages :

هو تأثير الفيروس على الخلايا خارج بيئتها لتشخيص العدوى الفيروسية

Cell culture	NO of sub passages	What kind of viruses their are	picture
Primary cell culture	1 or 2(it lasts for very short period)	Usually RNA	
Diploid cell culture (semi – continuous)	20-50(it lasts for few weeks)	Usually DNA	
Continuous cell line	Indefinite		

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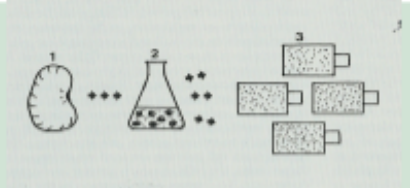
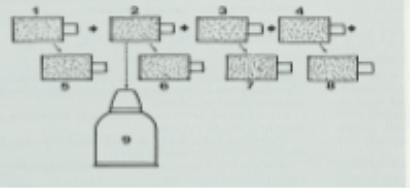
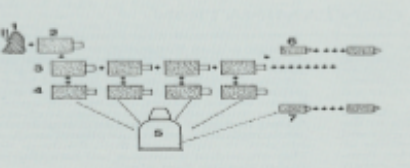
Note :there are Variation of Sensitivity of cell cultures to infection by viruses.
يعني الخلايا التي في العبوات تختلف حساسيتها تجاه الفيروس وهذا الشيء الذي يحددها
Primary or diploid or continues



What is meant by passage in cell culture?
In biology, a subculture is a new **cell** or microbiological **culture** made by transferring some or all **cells** from a previous **culture** to fresh growth medium. This action is called subculturing or passaging the **cells**. Subculture is used to prolong the life and/or expand the number of **cells** or microorganisms in the **culture**

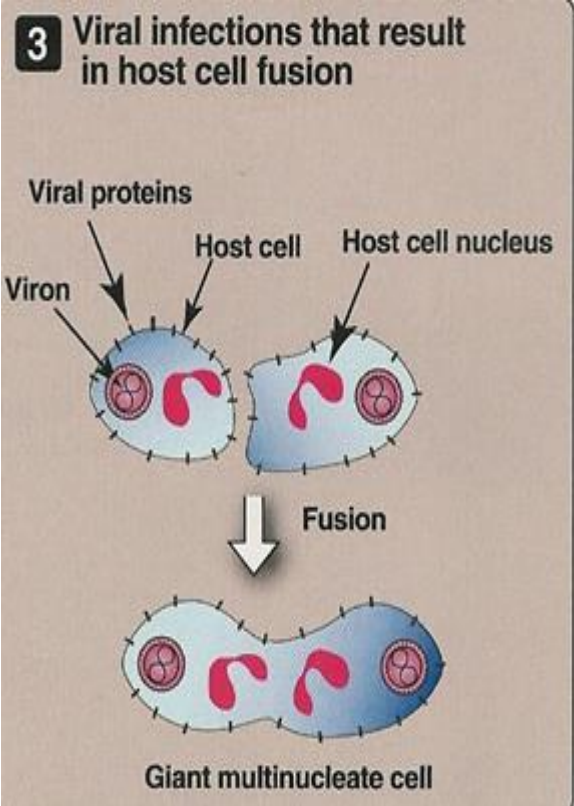
The term **passage number** refers to the **number** of times that a **cell** population has been removed from the **culture** vessel and undergone a subculture (**passage**) process, in order to keep the **cells** at a sufficiently low density to stimulate further growth.

نفس السلايد الي قبل + شرح

Cell culture	NO of sub passages	What kind of viruses their are	picture
Primary cell culture	1 or 2(it lasts for very short period)	Usually RNA	
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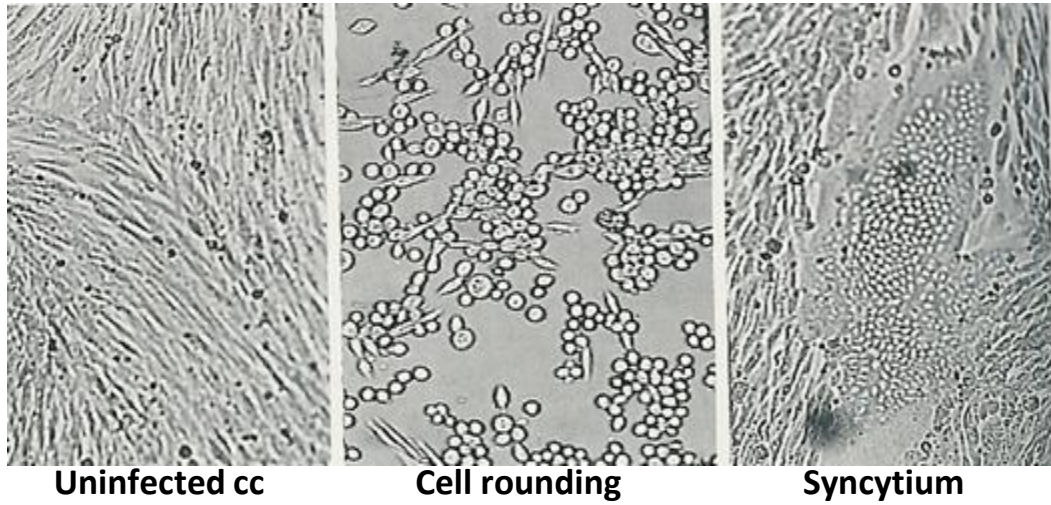


Cell culture

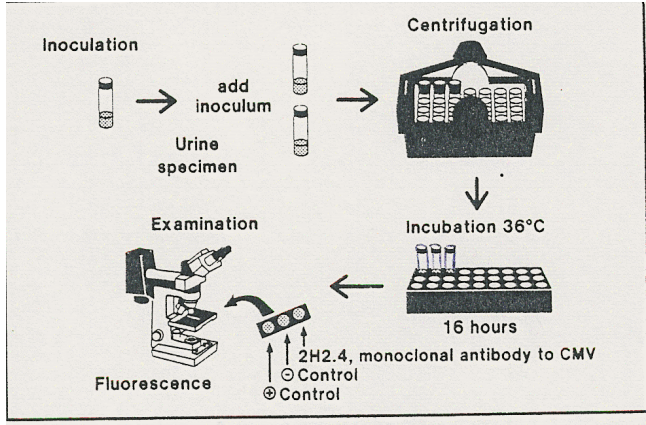


❖ Detection of viral growth:

1. By the **cytopathic effects** The affected cell will have "Rounding, shrinkage, aggregation, Syncytium(giant multinucleate cell) and lose of adherence.
2. Other
3. By Immunofluorescence (IF)



❖ **Rapid culture technique :**
Shell viral assay It detect **viral antigens** within 1-3 days



Problems with cell culture :

- Long incubation (up to 5 days) can be solved with rapid culture technique
- Sensitivity is variable
- Susceptible to bacterial contamination (عرضه للتلوث البكتيري)
- Some Viruses do not grow in cell culture ex. HCV

Laboratory Diagnosis of Viral Infections

-3-SEROLOGICAL TEST:



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To locate antibody or antigen:

- Immunofluorescence (IF)
- Enzyme-linked immunosorbent assay (ELISA)
- Molecular test

Uses the same technique

1- ELISA

2- IF

To find either antigen or antibody!

Antigen detection

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

sample	virus	test
Skin scrapings	HSV	IF
Blood	(HBV)hepatitis B virus (HBsAg)	ELISA

Laboratory Diagnosis of Viral Infections -3-SEROLOGICAL TEST:

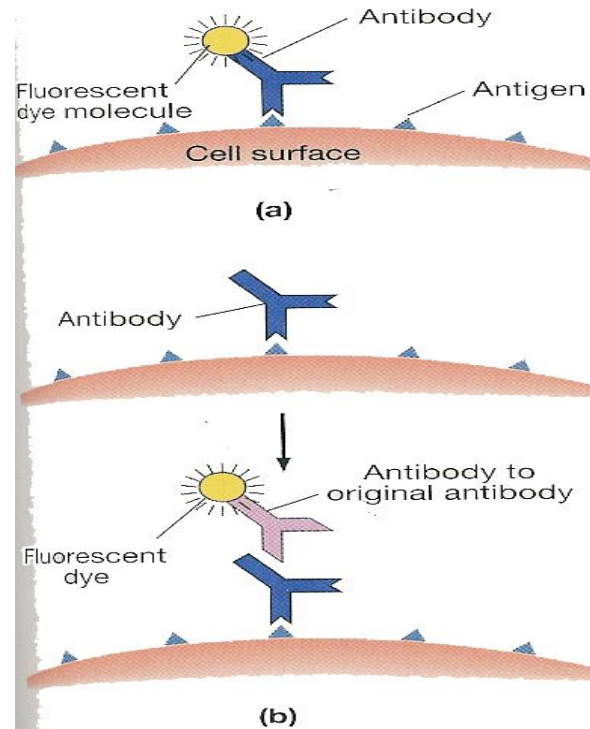


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Immunofluorescence (IF)

using a fluorescent dye And can be viewed
by fluorescence microscope .

- **Direct Ag** detection; Sample (Antigen)
- **Indirect Ab** detection; Sample (Antibody)



Laboratory Diagnosis of Viral Infections

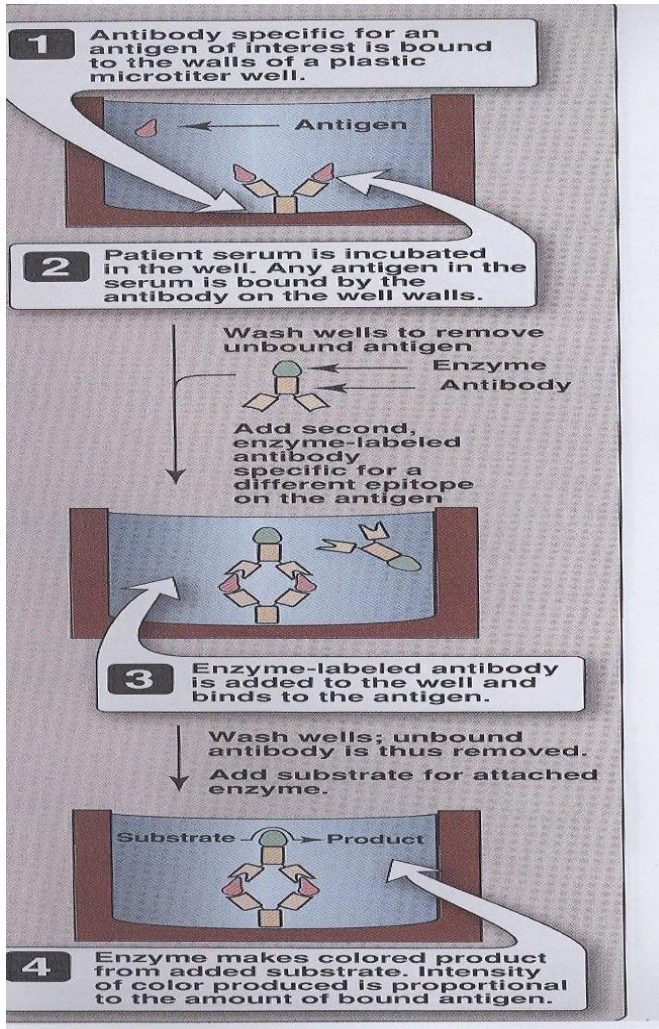
-3-SEROLOGICAL TEST:



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-Direct ELISA for Antigens detection

or specific IgM, IgG

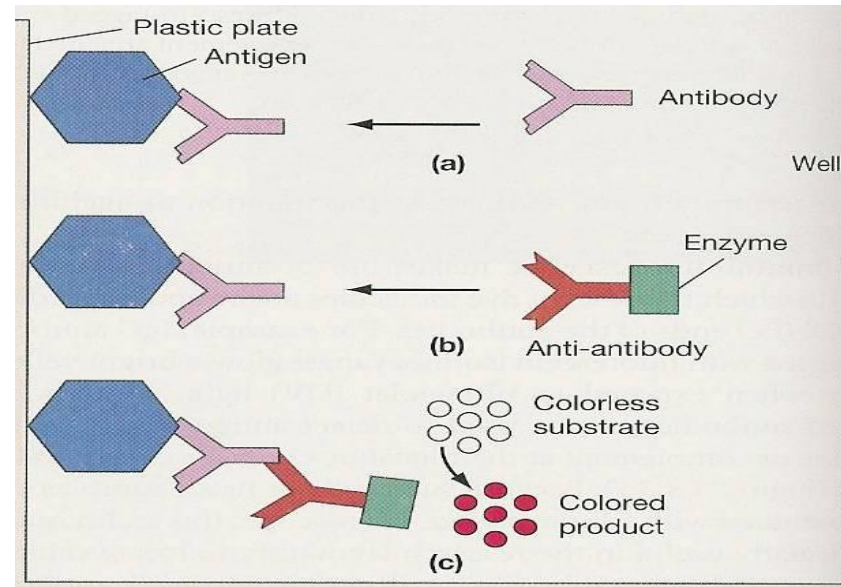


enzyme-linked immunosorbent assay (ELISA)

ملاحظة : الطرق أحيانا تكون مباشرة وغير مباشرة . اذا كانت غير مباشرة فهي عن طريق الأنتي بديز واذا مباشرة على طول تكون عن طريق الأنتي جين

Sample is viewed using spectrometer.

-Indirect ELISA for Antibodies detection ; colored wells indicate reactivity

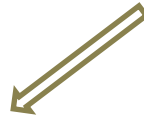
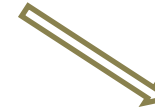


Laboratory Diagnosis of Viral Infections



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Molecular test



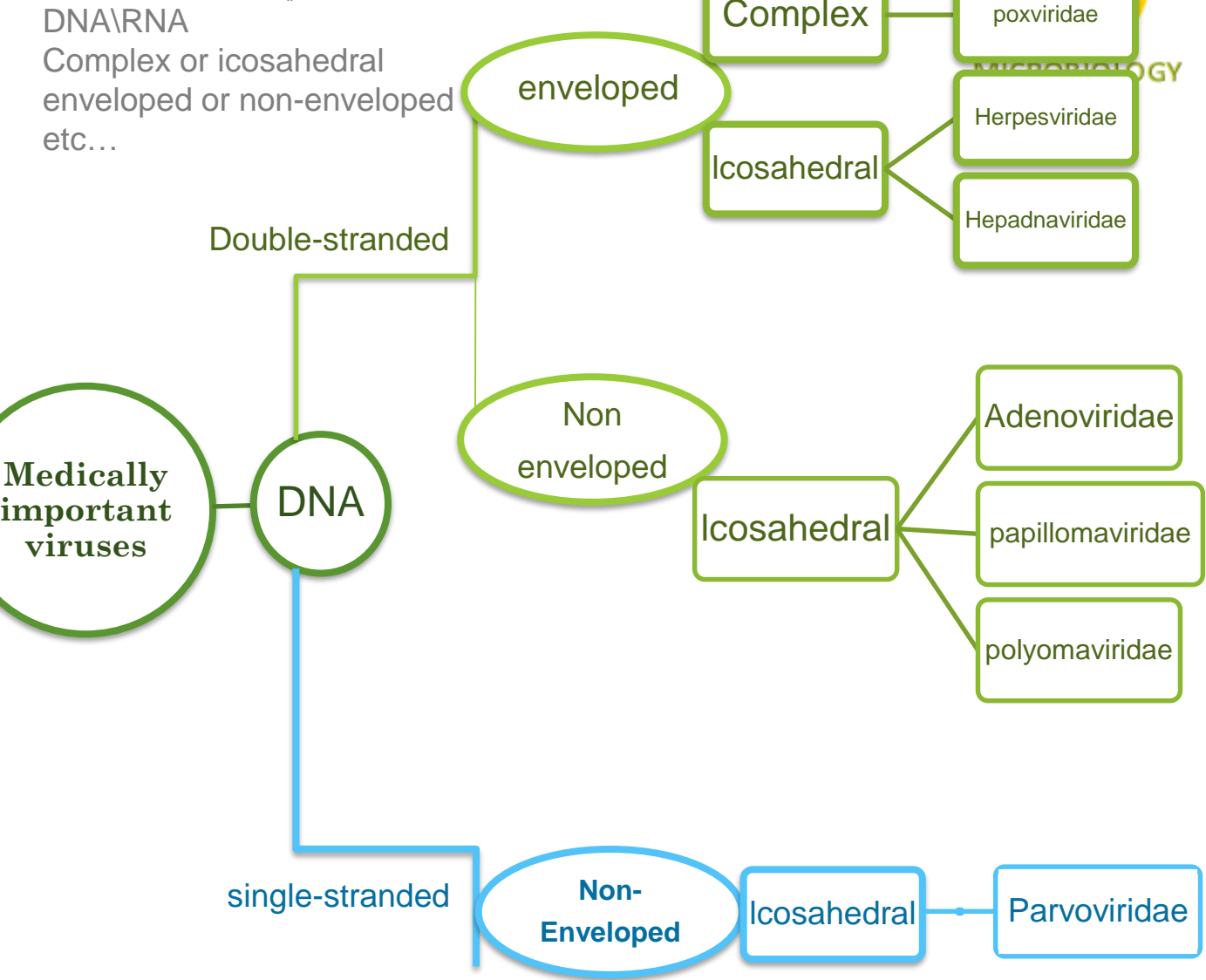
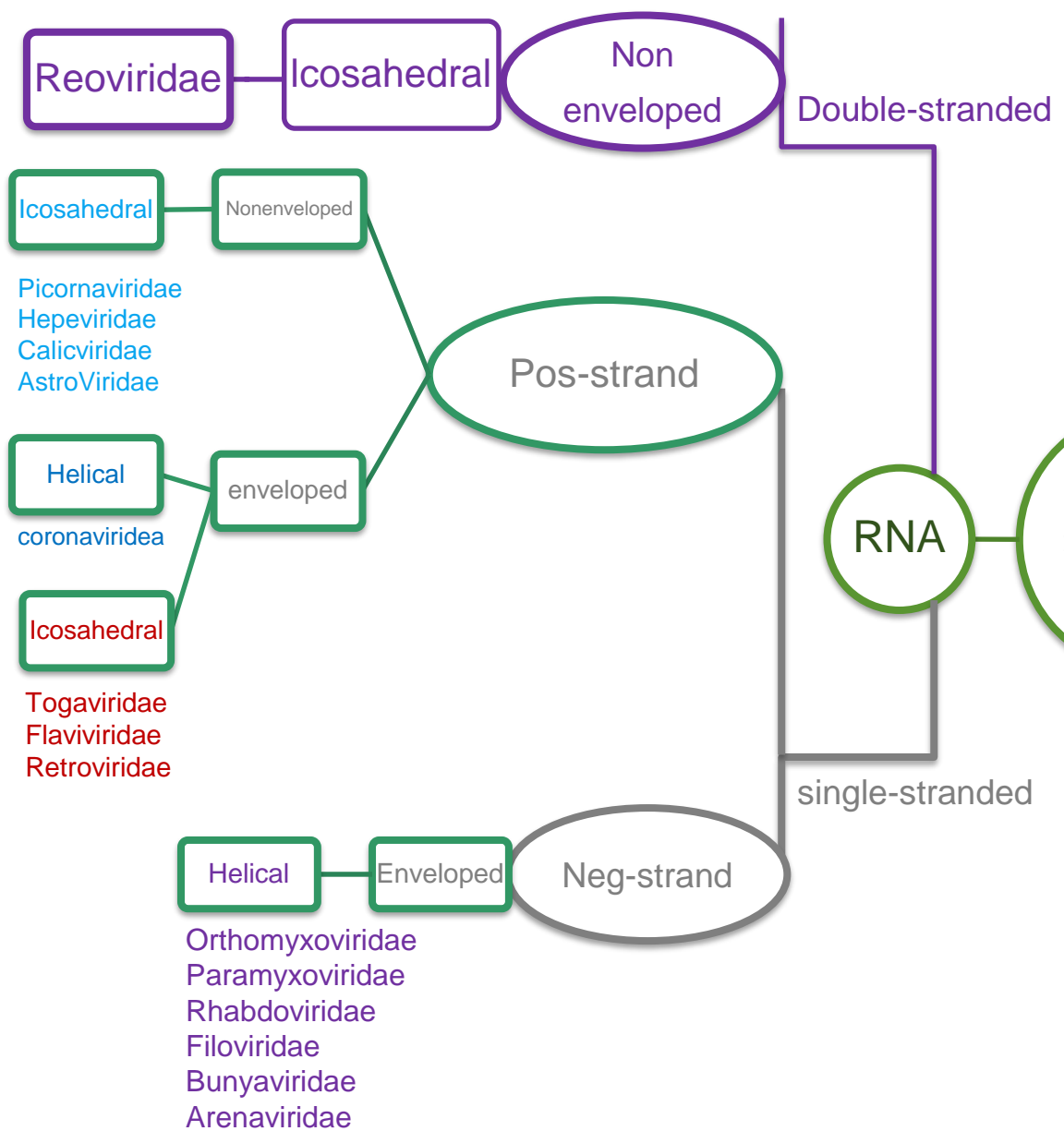
Used for Diagnosis
Only way to **measure viral load**
كمية الفيروسات

❖ **Polymerase chain reaction (PCR):**
- NA Amplification technique.
-Viral genome

❖ **Uses:**
❖ -Diagnosis
-Monitoring response to treatment

Molecular diagnostics is a collection of techniques used to analyze biological markers in the genome and proteome—the individual's genetic code and how their cells express their genes as proteins—by applying **molecular** biology to medical **testing**.

اسامي الفيروسات غير مطلوبه مننا في الوقت الحالي بس اعرفوا التقسيمات.
DNA\RNA
Complex or icosahedral enveloped or non-enveloped etc...





Quiz and references

1- Viruses contain :

- A) RNA b)DNA c)RNA or DNA d) both RNA and DNA

2- Direct ELISA detection for which of the following:

- A) RNA b) DNA c) antibody d)antigen

3- All viruses are haploid except :

- A)retroviruses b)phage c)herpesviridae d)parvoviruses

4- The viral envelope is composed from :

- A) lipoprotein b)polysaccharides c) proteins d)glycoprotein



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KING SAUD UNIVERSITY

لايقوى الإنسان في الحياة على هذه الأرض من دون أن يعاونه النَّاس ويقفوا معه.



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Team members:

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For any corrections, suggestions or any useful information please
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