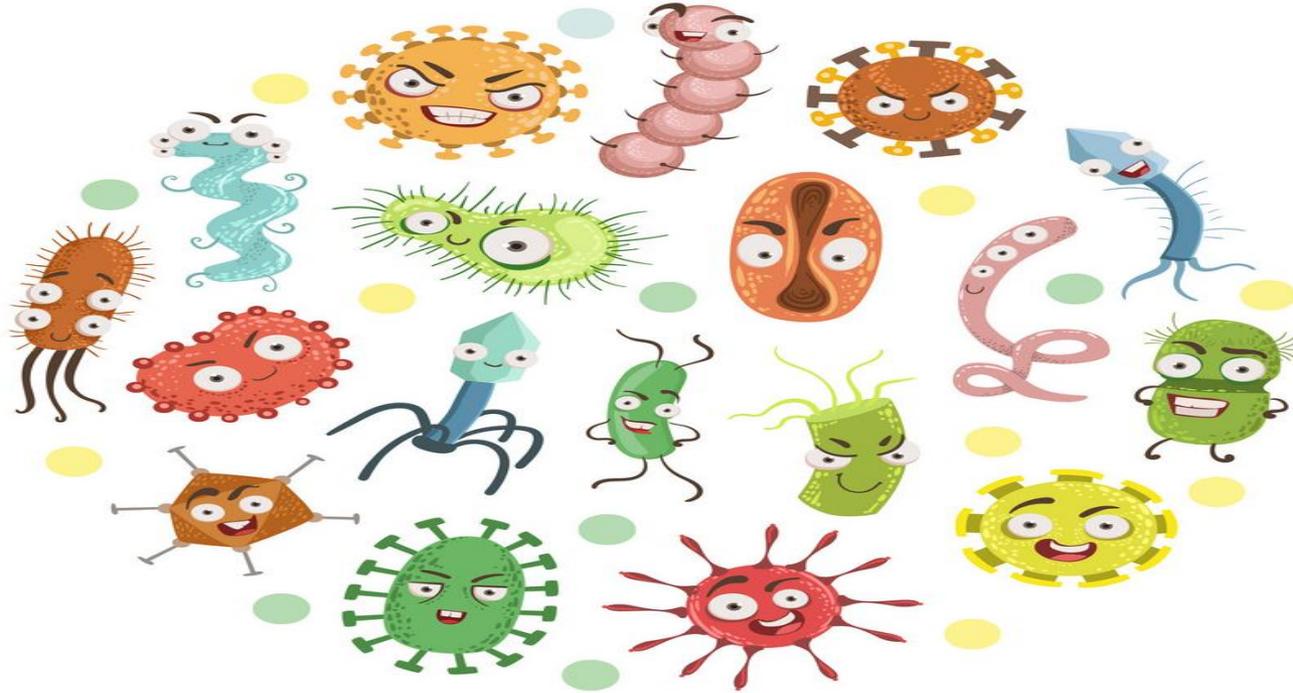




BIOLOGY AND DIVERSITY OF VIRUSES, BACTERIA AND FUNGI (PAPER CODE: BOT 501)



By

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OBJECTIVES

The main objective of the present lecture is to cover all the topics of 5 units under Block -1 in Paper code BOT 501 and to make them easy to understand and interesting for our students/learners.

BLOCK – I : VIRUSES

Unit –1 : General Characters and Classification of Viruses

Unit –2 : Chemistry and Ultrastructure of Viruses

Unit –3 : Isolation and Purification of Viruses

Unit –4 : Replication and Transmission of Viruses

Unit –5 : General Account of Plant, Animal and Human Viral Disease

CONTENT

- Introduction of viruses
- Origin of viruses
- History of viruses
- Classification
- Ultrastructure of viruses
- Chemical composition viruses
- Isolation and purification of viruses
- Replication of viruses
- Transmission of viruses
- General account of plant, animal and human viral diseases
- Key points
- Terminology
- Assessment Questions
- Bibliography

WHAT ARE THE VIRUSES ???

- ❖ Viruses are simple and acellular infectious agents.

Or

- ❖ Viruses are infectious agents having both the characteristics of living and nonliving.

Or

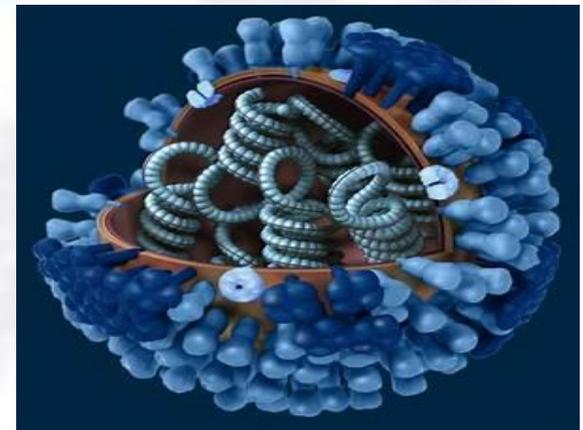
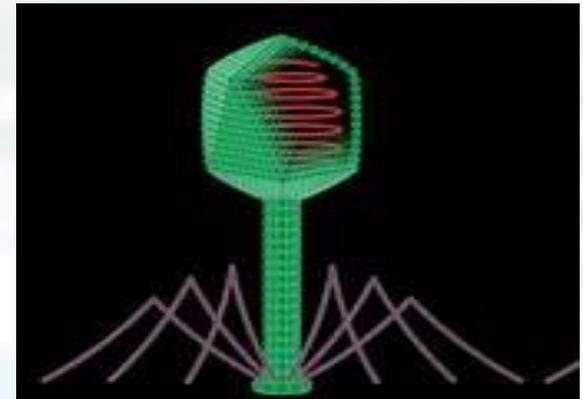
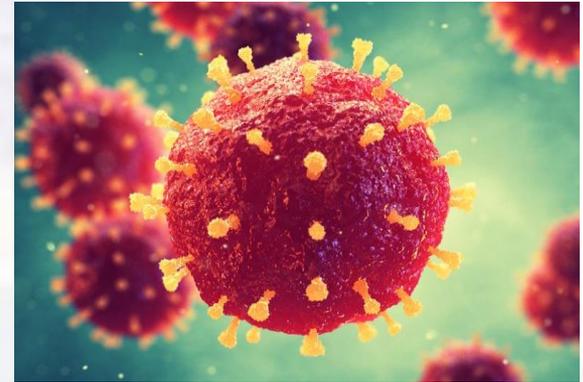
- ❖ Viruses are microscopic obligate cellular parasites, generally much smaller than bacteria. They lack the capacity to thrive and reproduce outside of a host body.

Or

- ❖ Viruses are infective agent that typically consists of a nucleic acid molecule in a protein coat, is too small to be seen by light microscopy, and is able to multiply only within the living cells of a host.

Or

- ❖ Viruses are the large group of submicroscopic infectious agents that are usually regarded as nonliving extremely complex molecules, that typically contain a protein coat surrounding an RNA or DNA core of genetic material but no semipermeable membrane, that are capable of growth and multiplication only in living cells, and that cause various important diseases in humans, animals, and plants.



ORIGIN OF VIRUSES

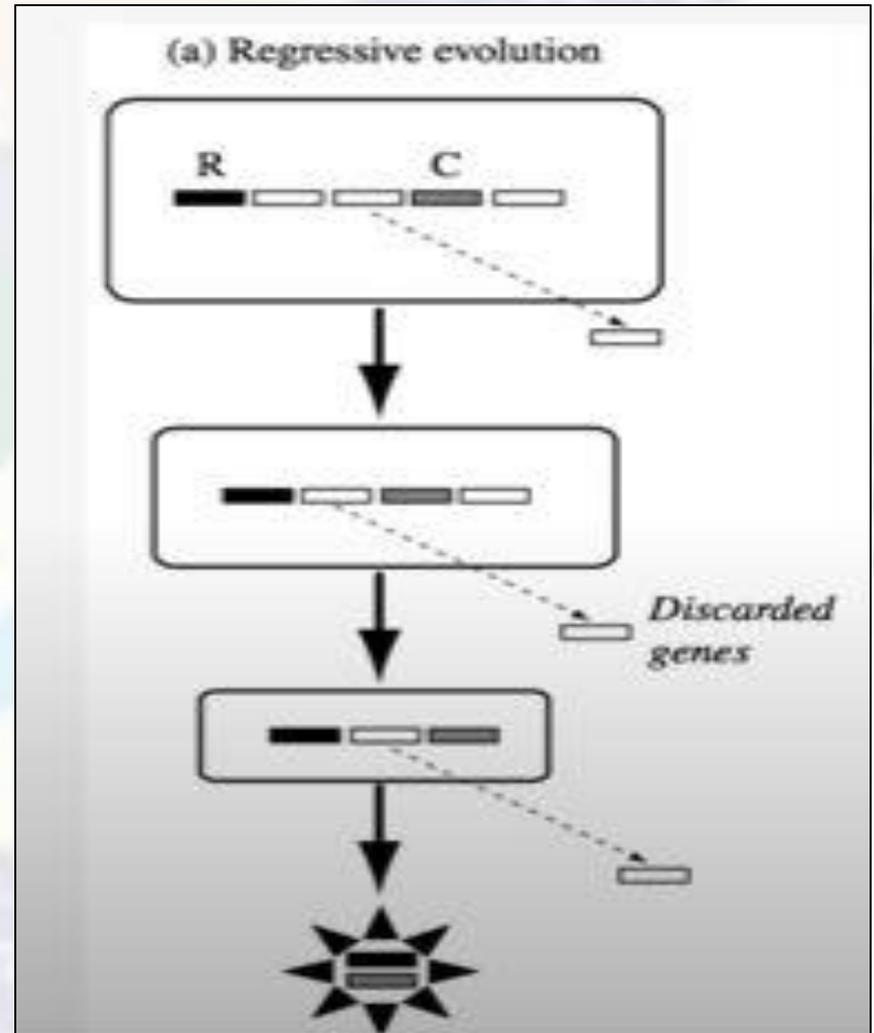
- ❖ The origin of viruses is unclear because they do not form fossils.
- ❖ There are three main hypotheses that aim to explain the origins of viruses:

I-Regressive Hypothesis

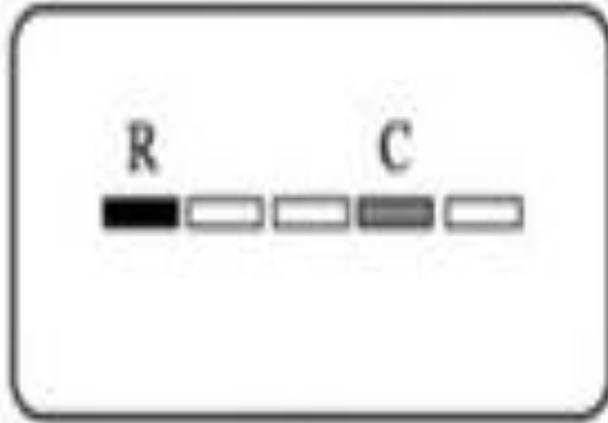
- This is also called the 'degeneracy hypothesis' or 'reduction hypothesis'.
- Viruses may have once been small cellular organisms that parasitized larger cells.
- Over time, genes not required by them and they become simpler organisms which are the viruses we see today.

Supporting Evidence:

Rickettsia and Chlamydia are living cells that, like viruses, can reproduce only inside host cells. They lend support to this hypothesis, as their dependence on parasitism is likely to have caused the loss of genes that enabled them to survive outside a cell.



(b) Escaped host gene



II- Escaped Gene Theory

- Some viruses may have evolved from bits of DNA or RNA that "escaped" from the genes of a larger organism.
- The escaped DNA could have come from plasmids (pieces of naked DNA that can move between cells) and ortransposons (molecules of DNA that replicate and move around to different positions within the genes of the cell).
- This is sometimes called the *vagrancy hypothesis* or the *escape hypothesis*.

Supporting Evidence:

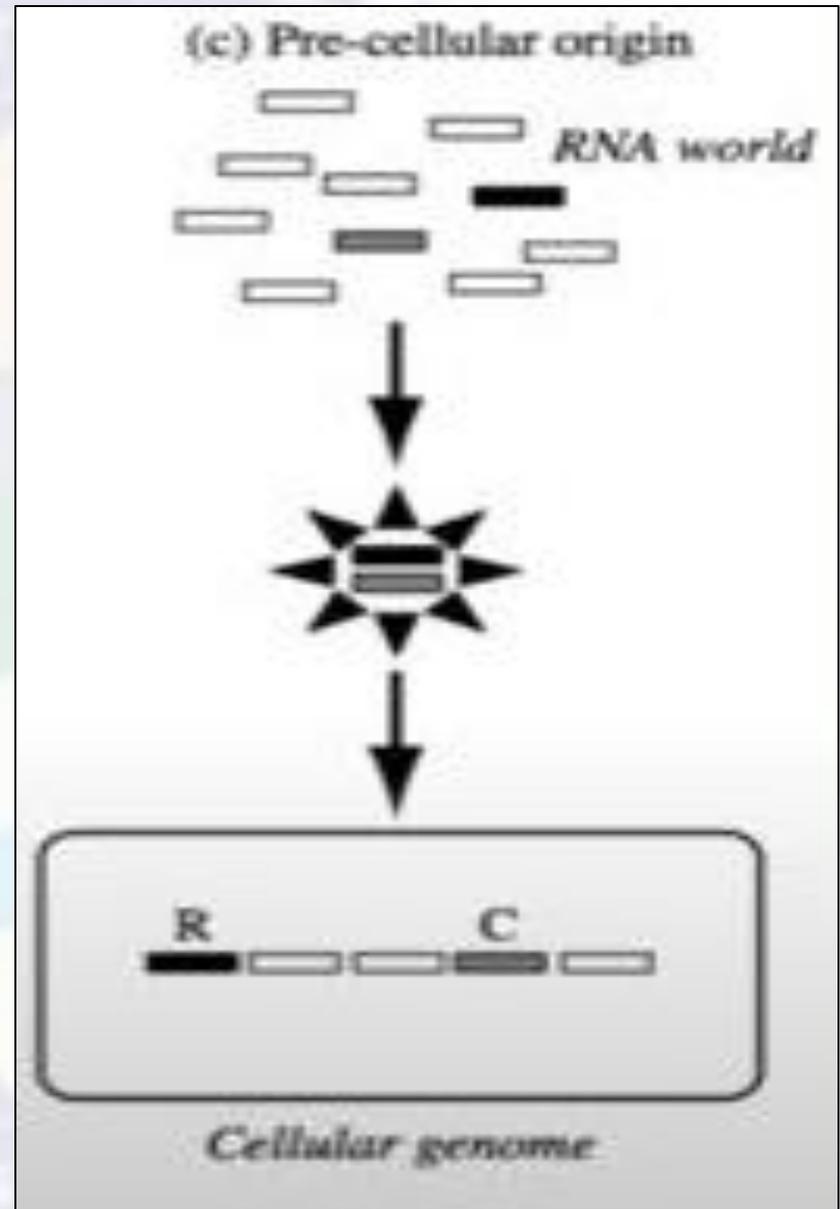
Once called "jumping genes", transposons are examples of mobile genetic elements and could be the origin of some viruses. They were discovered in maize by Barbara McClintock in 1950.

III-Co-evolution Hypothesis

- This is also called the *virus-first hypothesis*.
- This hypothesis proposes that viruses may have evolved from complex molecules of protein and nucleic acid at the same time as cells first appeared on Earth and would have been dependent on cellular life for billions of years.

Supporting Evidence:

- Viruses with similar viral machinery are present in the three groups of life; Bacteria, Archaeobacteria and Eukaryotes.
- DNA viruses are remnants of the first DNA replicators, while the retroviruses could be descendants of the first molecules that were able to make the transition between RNA and DNA.

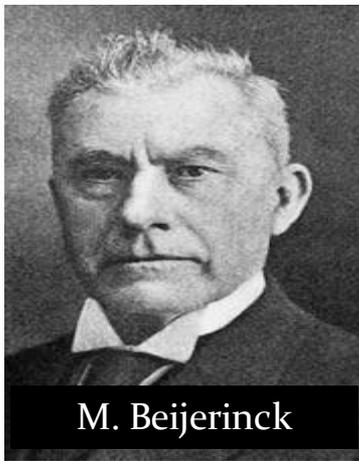


HISTORY OF VIRUSES

- ❖ The branch of science which deals with the study of viruses is called **virology**.
- ❖ The term “virus” is derived from the Latin word *vīrus* referring to poison and other noxious liquids.
- ❖ Viruses can infect all types of life forms including multicellular organisms to unicellular organisms.
- ❖ Many of human, animal and plant diseases are caused by them. Even the recently appeared pandemic “COVID-19” is also caused by virus.



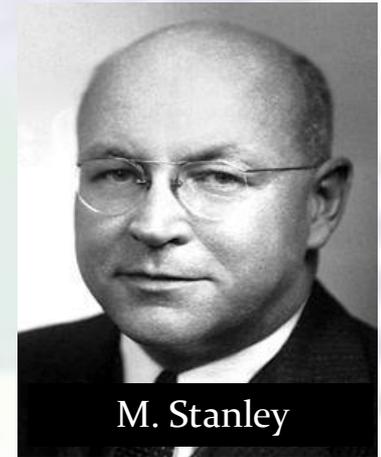
- ❖ In 1884, the French microbiologist Charles Chamberland invented a filter (known today as the Chamberland filter or the Pasteur-Chamberland filter) with pores smaller than bacteria. Thus, he could pass a solution containing bacteria through the filter and completely remove them.
- ❖ In 1892, the Russian biologist Dmitri Ivanovsky first discovered virus in an infected tobacco plant.



M. Beijerinck

- ❖ In 1898, the Dutch microbiologist Martinus Beijerinck observed that these “agents” multiplied only in cells that were dividing. He called it a *contagium vivum fluidum* (soluble living germ) and re-introduced the word *virus*.
- ❖ The first images of viruses were obtained upon the invention of electron microscopy in 1931 by the German engineers Ernst Ruska and Max Knoll.

- ❖ The Tobacco Mosaic Virus was the first to be crystallized by Wendell Meredith Stanley in 1935 and its structure could therefore be explained in detail.
- ❖ By the end of the 19th century, viruses were defined in terms of their infectivity, ability to be filtered, and their requirement for living hosts. Viruses had been grown only in plants and animals.



M. Stanley



F. Twort

- ❖ In the early 20th century, the English bacteriologist Frederick Twort discovered a group of viruses that infect bacteria, now called bacteriophages.
- ❖ Microbiologist Félix d'Herelle described viruses that, when added to bacteria on an agar plate, would produce areas of dead bacteria.
- ❖ The second half of the 20th century was the golden age of virus discovery and most of the over 2,000 recognized species of animal, plant, and bacterial viruses were discovered during these years.

CHARACTERS AND PROPERTIES OF VIRUSES

- ❖ Viruses are a cellular, non-cytoplasmic infectious agents. Therefore, a unit of virus is referred to as 'a virus particle' rather than 'a virus cell'.
- ❖ They are smaller than bacteria and can pass through bacteriological filter.
- ❖ They are consisting mainly of a nucleic acid surrounded by a protein envelope called capsid.
- ❖ They are devoid of the sophisticated enzymatic and biosynthetic machinery essential for independent activities of cellular life. Therefore, they can grow only inside suitable living cells.
- ❖ These viruses do not grow, neither respire nor metabolize, but they reproduce.
- ❖ Viruses may even be crystallized much like molecules although some kind of viruses can only be purified but not crystallized .
- ❖ A virus cannot contain both DNA and RNA. Therefore, virus is called either 'DNA virus' or 'RNA virus' depending on whether it contains the nucleic acid DNA or RNA.
- ❖ Viruses are transmissible from disease to healthy organisms.
- ❖ All viruses are obligate parasites and can multiply only within the living host cells.
- ❖ Viruses are host specific that they infect only a single species and definite cells of the host.
- ❖ They are highly resistant to germicides and extremes of physical conditions.
- ❖ Viruses are called connective link between living and non living.

Why do we consider viruses as a connective link between living and non living ?????

Viruses are living:

- ✓ They possess genetic material i.e., either DNA or RNA.
- ✓ They can undergo mutation.
- ✓ They show irritability.
- ✓ They are capable to reproduce
- ✓ They can increase their number.
- ✓ They can be transmitted from one host to another.
- ✓ They react to heat, chemicals and radiations.
- ✓ These develop resistant to antibiotics.

Virus

Viruses are non-living:

- ✓ They can be crystallized.
- ✓ They are inert outside the host.
- ✓ Lack cell membrane and cell wall.
- ✓ Lack of cytoplasm and organelles.
- ✓ They do not show cell division.
- ✓ They can't grow in size, shape.
- ✓ They don't possess sort of nutrients.
- ✓ They don't respire and excrete.
- ✓ Don't undergo their own metabolism.
- ✓ Lack any energy producing system.

CLASSIFICATION OF VIRUSES

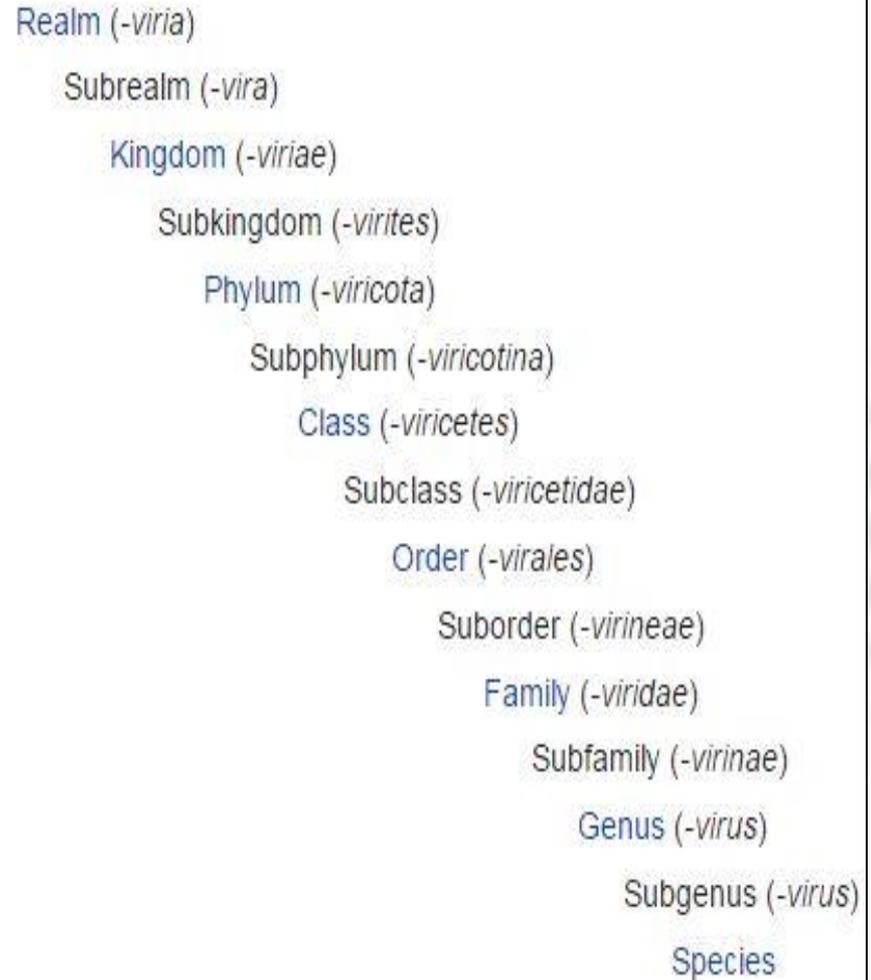
- ❖ Virus classification is the process of naming viruses and placing them into a taxonomic system.
- ❖ Viruses do not fit into the established biological classification of cellular organisms. This is mainly due to pseudo-living nature of viruses.
- ❖ Initially, on the basis of their host range, clinical, epidemiological and pathological symptoms, viruses were classified into the following four groups:
 - Plant viruses : This group includes only plants virus
 - Invertebrate viruses : This group includes only invertebrates virus
 - Vertebrate Viruses : This group includes viruses infecting vertebrate.
 - Dual-host viruses : Infects two different hosts mentioned above.

1. Holmes Classification

- ❖ Holmes (1948) included all viruses in a single order Virales which were divided into three sub-orders:
 - Phagineae : This sub-order includes viruses infecting bacteria *i.e.*, bacteriophage.
 - Phytophagineae : It includes viruses infecting plants.
 - Zoophagineae : It includes viruses infecting animals.

2. ICTV Classification

- ❖ The International Committee on Taxonomy of Viruses began to devise and implement rules for the naming and classification of viruses early in the 1970s.
- ❖ The ICTV is the only body charged by the International Union of Microbiological Societies with the task of developing, refining, and maintaining a universal virus taxonomy.
- ❖ The system shares many features with the classification system of cellular organisms, such as taxon structure.
- ❖ However, some differences exist, such as the universal use of italics for all taxonomic names, unlike in the International Code of Nomenclature for algae, fungi, and plants and International Code of Zoological Nomenclature.
- ❖ Viral classification starts at the level of realm and continues as follows, with the taxonomic suffixes in parentheses.



- ❖ Species names often take the form of *[Disease] virus*, particularly for higher plants and animals.

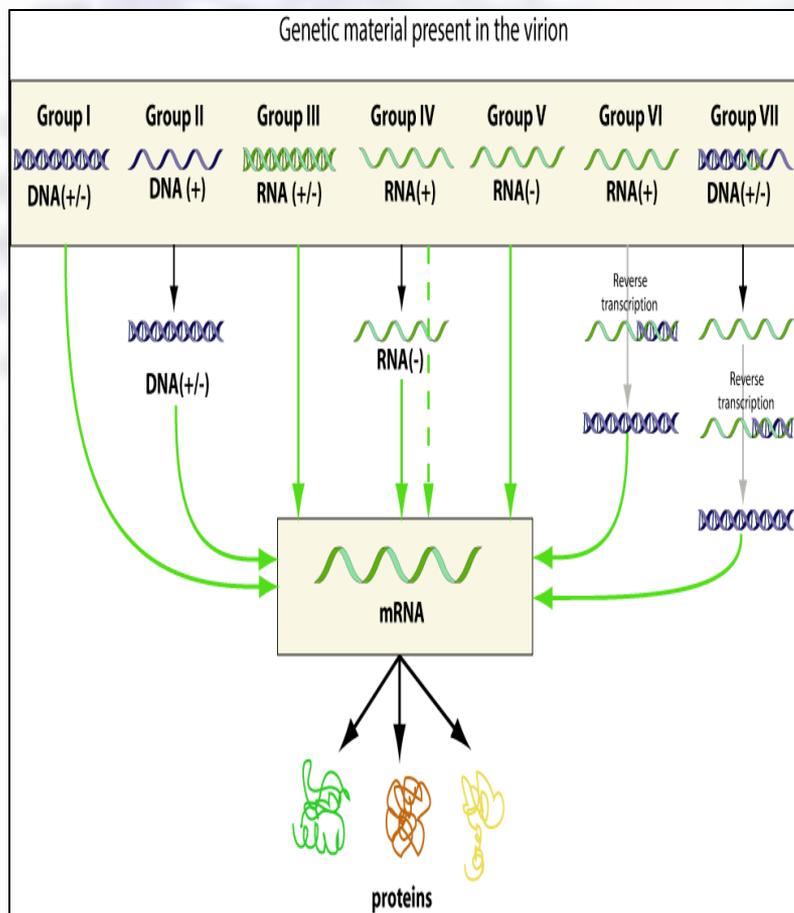
- ❖ As of 2019, all levels of taxa except subrealm, subkingdom, and subclass are used.
- ❖ Four realms, one *incertae sedis* order, 24 *incertae sedis* families, and three *incertae sedis* genera are recognized (Virus Taxonomy: 2019 Release):
- ❖ **Realms:** *Duplodnaviria*, *Monodnaviria*, *Riboviria*, and *Varidnaviria*
- ❖ ***Incertae sedis* order:** *Ligamenvirales*
- ❖ ***Incertae sedis* families:**
 - *Alphasatellitidae*
 - *Clavaviridae*
 - *Hytrosaviridae*
 - *Portogloboviridae*
 - *Ampullaviridae*
 - *Finnlakeviridae*
 - *Nimaviridae*
 - *Pospiviroidae*
 - *Anelloviridae*
 - *Fuselloviridae*
 - *Nudiviridae*
 - *Spiraviridae*
 - *Avsunviroidae*
 - *Globuloviridae*
 - *Ovaliviridae*
 - *Thaspiviridae*
 - *Baculoviridae*
 - *Guttaviridae*
 - *Plasmaviridae*
 - *Tolecusatellitidae*
 - *Bicaudaviridae*
 - *Halspiviridae*
 - *Polydnaviridae*
 - *Tristromaviridae*
- ❖ ***Incertae sedis* genera:** *Deltavirus*, *Dinodnavirus*, *Rhizidiovirus*

3. Structure Based Classification

- ❖ It has been suggested that similarity in virion assembly and structure observed for certain viral groups infecting hosts from different domains of life (e.g., bacterial tectiviruses and eukaryotic adenoviruses or prokaryotic Caudovirales and eukaryotic herpesviruses) reflects an evolutionary relationship between these viruses (Bamford, 2003).
- ❖ Therefore, structural relationship between viruses has been suggested to be used as a basis for defining higher-level taxa – structure-based viral lineages – that could complement the existing ICTV classification scheme (Krupovič and Bamford, 2010).

4. Baltimore Classification

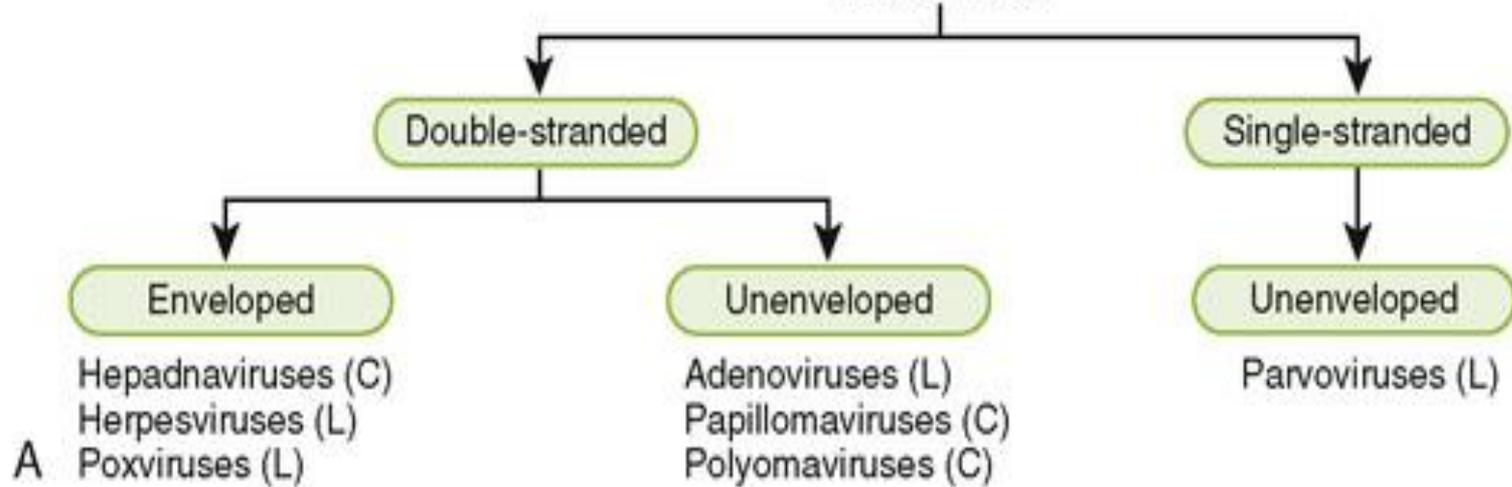
- ❖ Baltimore classification (first defined in 1971) is a classification system that places viruses into one of seven groups depending on a combination of their nucleic acid (DNA or RNA), strandedness (single-stranded or double-stranded), sense, and method of replication.
- ❖ Viruses can be placed in one of the seven following groups:



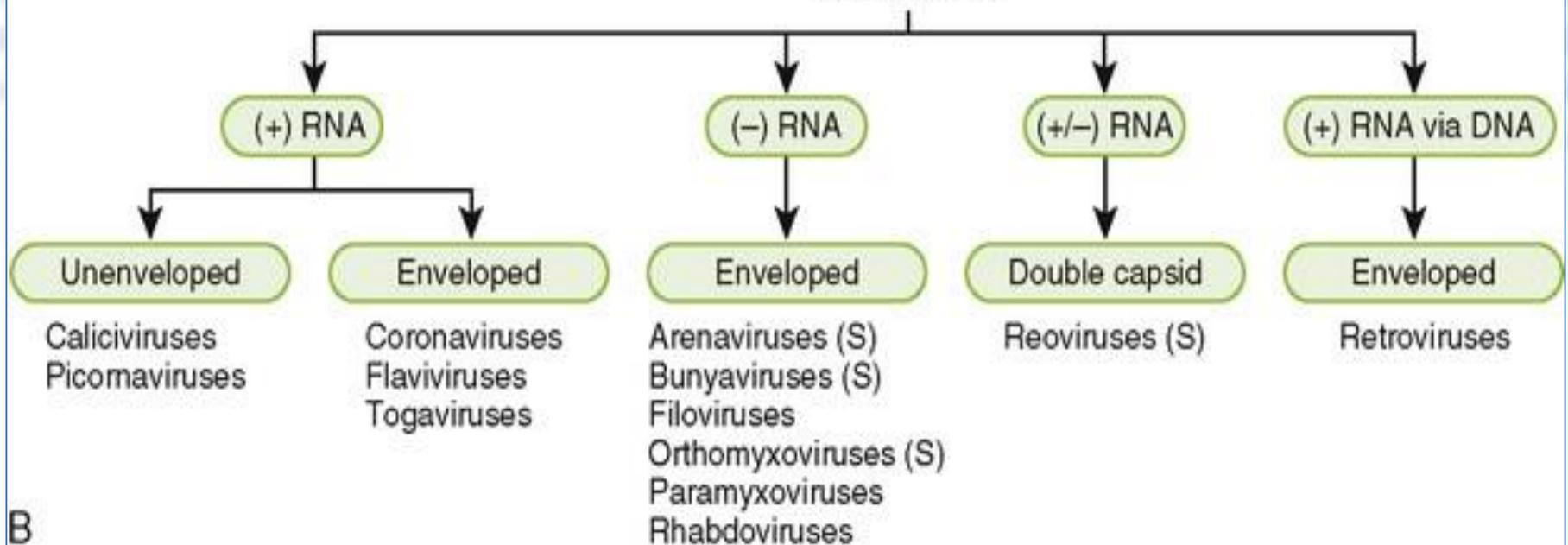
- Group I ds-DNA viruses
(e.g. Adenoviruses, Herpesviruses, etc)
- Group II ss-DNA viruses
(Parvoviruses)
- Group III ds- RNA viruses
(e.g. Reoviruses)
- Group IV Positive-sense ss- RNA viruses
(Coronaviruses, Picornaviruses, etc)
- Group V Negative-sense ss-RNA viruses
(e.g. Orthomyxoviruses, Rhabdoviruses)
- Group VI Reverse transcribing diploid ss-RNA viruses
(e.g. Retroviruses)
- Group VII Reverse transcribing circular ds-DNA viruses
(e.g. Hepadnaviruses)

ds=double-stranded, single-stranded=ss, double-stranded=ds,
Group I & II are DNA virus, group III, IV & V are RNA virus,
Group VI & VII are Reverse transcribing virus

DNA viruses



RNA viruses

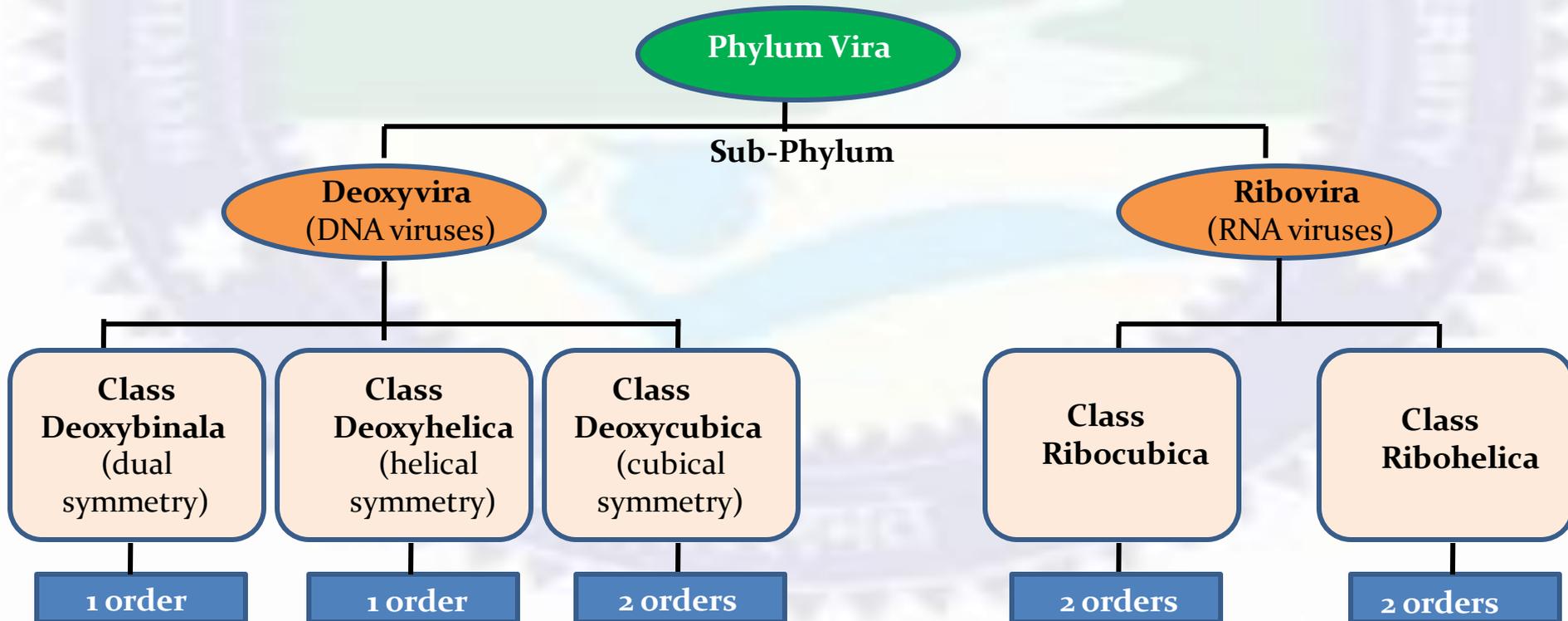


5. LHT System of Virus Classification

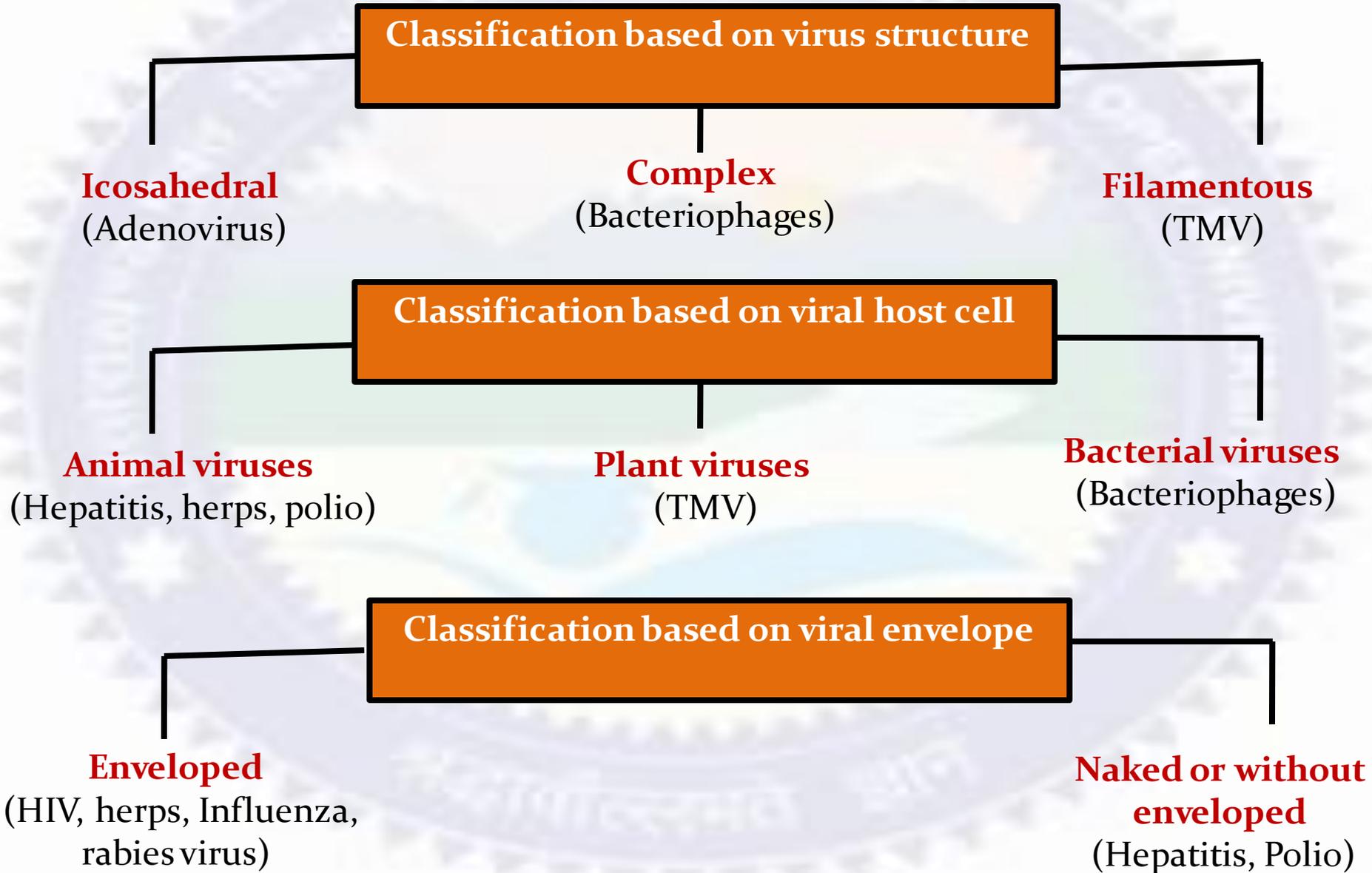
❖ The LHT System of virus classification is based on:

- Chemical and physical characters like nucleic acid (DNA or RNA),
- Symmetry (helical or icosahedral or complex),
- Presence of envelope,
- Diameter of capsid,
- Number of capsomers.

❖ This classification was approved by the Provisional Committee on Nomenclature of Virus (PNVC) of the International Association of Microbiological Societies (1962). It is as follow:



6. Virus Classification based on various properties



Phylum Vira (divided into 2 subphyla)

Subphylum Deoxyvira (DNA viruses)

Class Deoxybinala (dual symmetry)

Order Urovirales

Family Phagoviridae

Class Deoxyhelica (helical symmetry)

Order Chitovirales

Family Poxviridae

Class Deoxycubica (cubical symmetry)

Order Pellovirales

Family Herpesviridae (162 capsomeres)

Order Haplovirales (no envelope)

Family Iridoviridae (812 capsomeres)

Family Adenoviridae (252 capsomeres)

Family Papiloviridae (72 capsomeres)

Family Paroviridae (32 capsomeres)

Family Microviridae (12 capsomeres)

Subphylum Ribovira (RNA viruses)

Class Ribocubica

Order Togovirales

Family Arboviridae

Order Tymovirales

Family Napoviridae

Family Reoviridae

Class Ribohelica

Order Sagovirales

Family Stomataviridae

Family Paramyxoviridae

Family Myxoviridae

Order Rhabdovirales

Suborder Flexiviridales

Family Mesoviridae

Family Peptoviridae

Suborder Rigidovirales

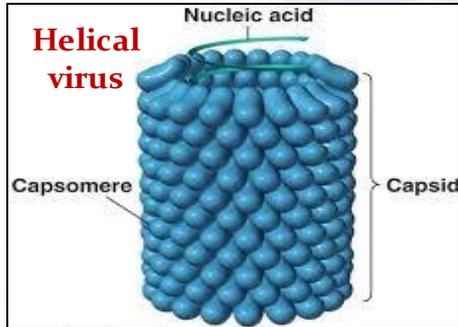
Family Pachyviridae

Family Protoviridae

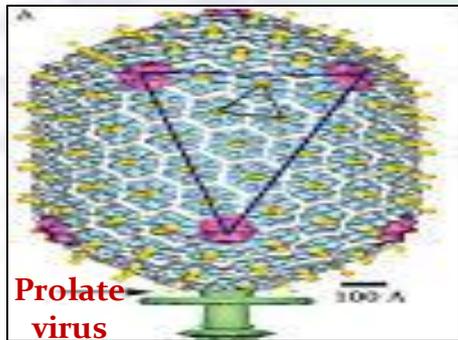
Family Polichoviridae

ULTRASTRUCTURE OF VIRUSES

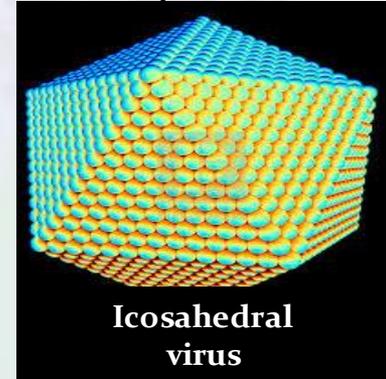
❖ Viruses may be classified into various **morphological types** on the basis of their capsid architecture



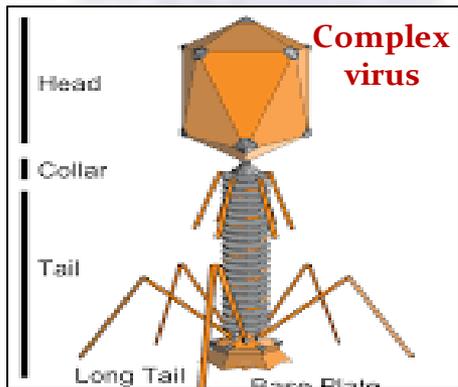
- **Helical viruses** : Composed of a single type of capsomere stacked around a central axis to form a helical structure, which may have a central cavity, or hollow tube. E.g: TMV



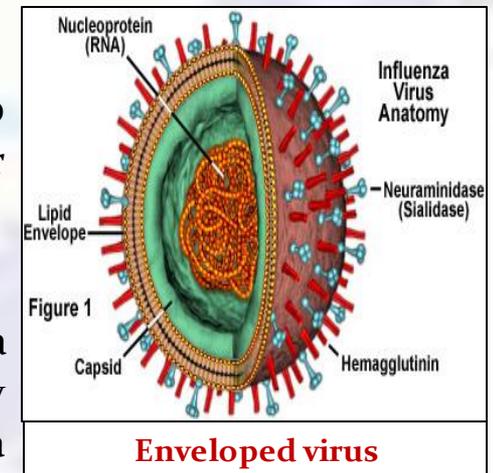
- **Icosahedral**: Most animal viruses are icosahedral or near-spherical with icosahedral symmetry. E.g: Adenovirus



- **Prolate**: This is an icosahedron elongated along one axis and is a common arrangement of the heads of bacteriophages.

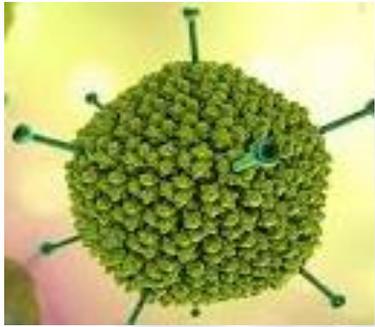


- **Enveloped viruses**: Some viruses envelop themselves in a modified outer lipid bilayer known as a viral envelope. E.g: HIV

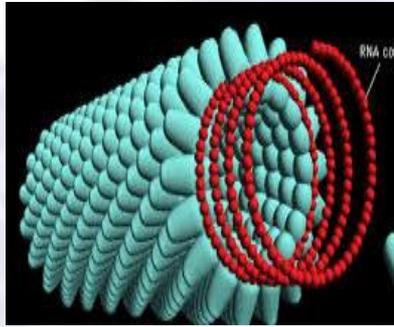


- **Complex viruses**: These viruses possess a capsid that is neither purely helical nor purely icosahedral, and that may possess extra structures such as protein tails or a complex outer wall. E.g: Bacteriophages

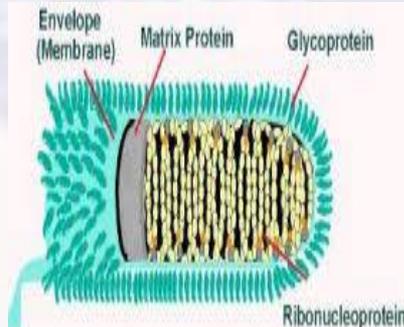
Shape of Viruses



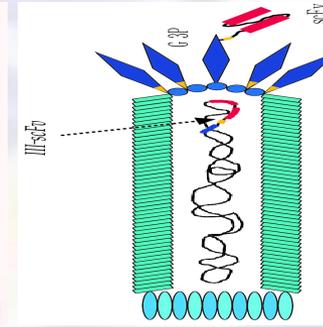
Spheroid virus
(E.g: Adenovirus)



Elongated virus
(E.g: Potato virus)



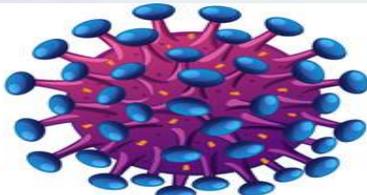
Bullet shaped
(E.g: Rabies virus)



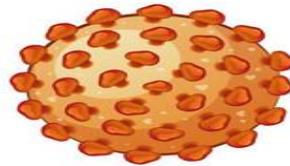
Filamentous virus
(E.g: Bacteriophage M 13)



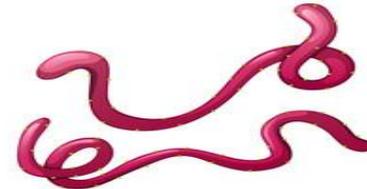
Asymmetrical viruses



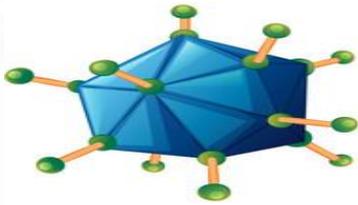
HIV



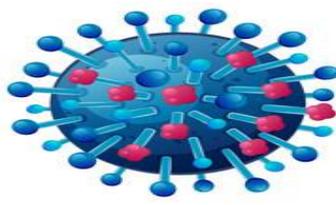
Hepatitis B



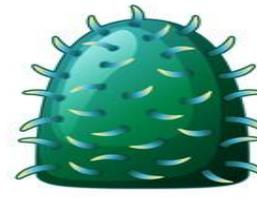
Ebola Virus



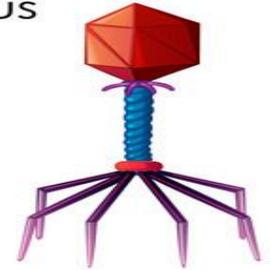
Adenovirus



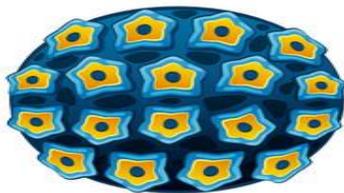
Influenza



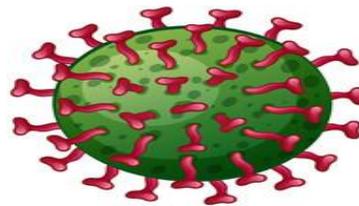
Rabies Virus



Bacteriophage



Papillomavirus



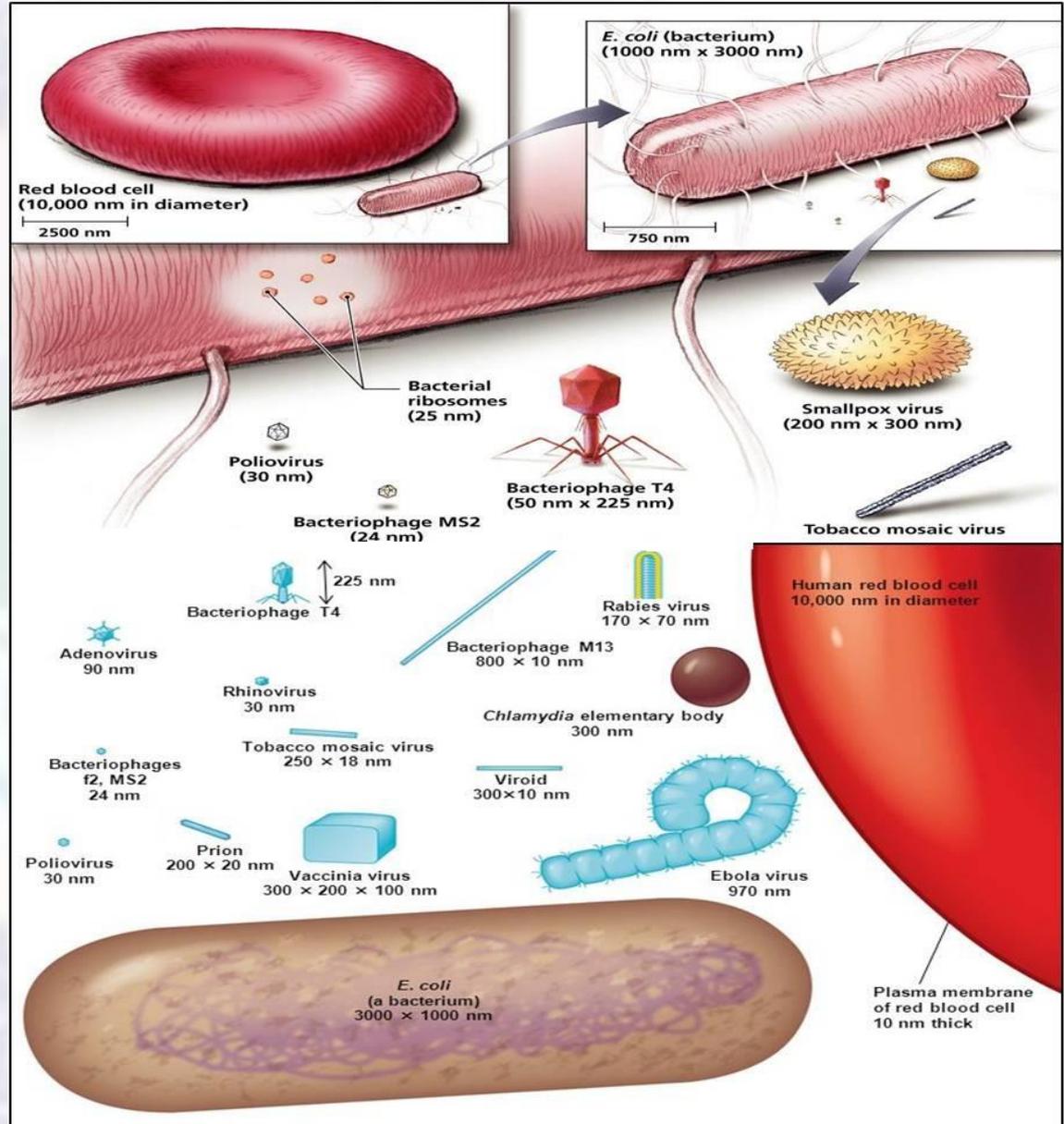
Rotavirus



Herpes Virus

Size of Viruses

- Viruses display a wide diversity of sizes.
- In general, viruses are much smaller than bacteria.
- Most viruses that have been studied have a diameter between 20 and 350 nanometres.
- They are smaller than bacteria.
- Some are slightly larger than protein and nucleic acid molecules
- Some are about of the same size (small pox virus) as the smallest bacterium and some virus (virus of lymphogranuloma, 300-400 um) are slightly larger than the smallest bacterium.



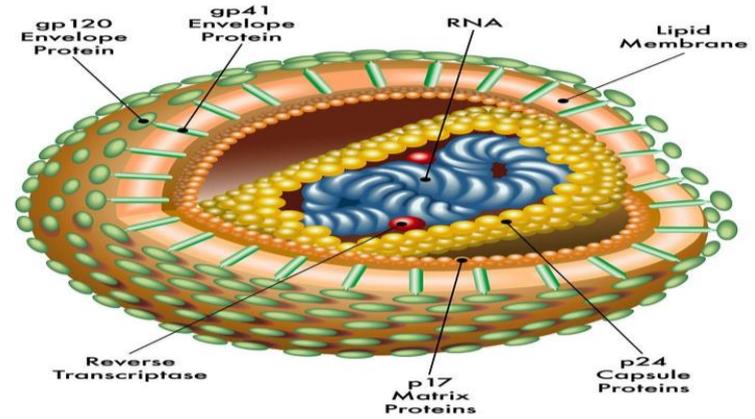
CHEMICAL COMPOSITION OF VIRUSES

1. Viral Protein: Proteins found in viruses may be grouped into the four categories:

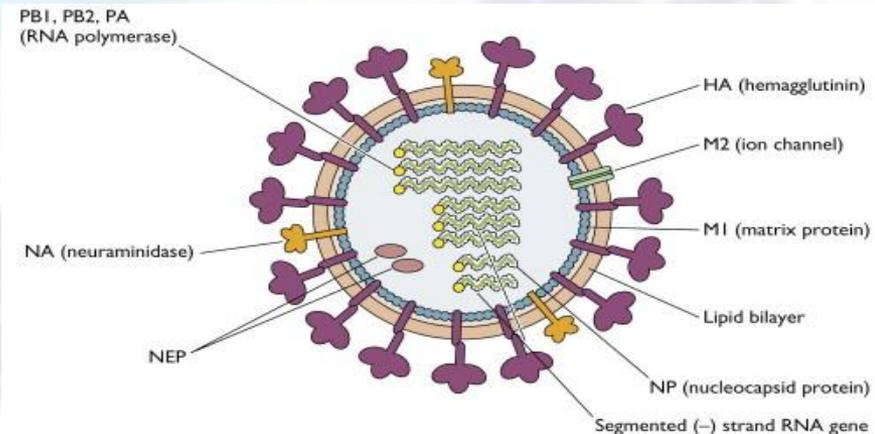
- **Envelope protein:** Enveloped viruses contain glycoprotein which differ from virus to virus.
- **Nucleocapside protein:** Viral capsids are made up totally of protein of identical subunits (promoters). E.g: capsids contain single type of protein in TMV.
- **Core protein:** Protein found in the nucleic acid is known as core protein.
- **Viral enzyme:** In animal viruses especially in the enveloped viruses, many virion specific enzymes have been characterized. E.g: RNase, reverse transcriptase in retrovirus.

2. Viral envelope: It is 10-15 μm thick, made up of protein, lipids and carbohydrate

- ❖ Lipid provide flexibility to the shape.
- ❖ The spikes attached to the outer surface of the envelope are made up of glycoproteins

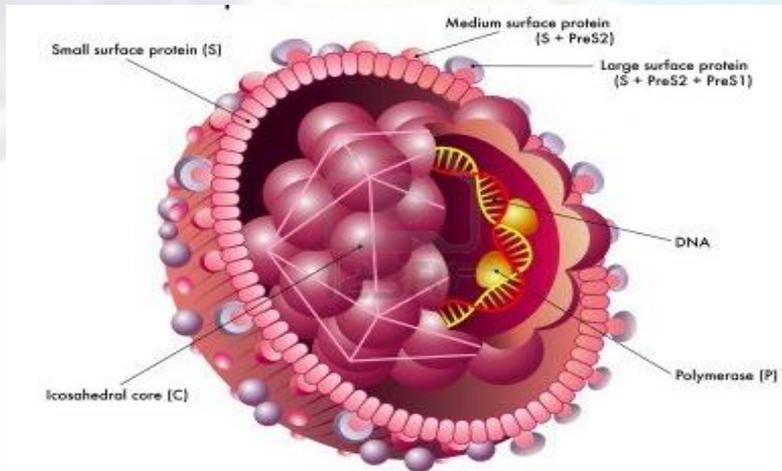


3. Viral carbohydrates: A substantial amount of carbohydrate specified by rather host cell (arbovirus) or viral genome (vaccinia virus) is found in viral envelope. For example galactose, mannose, glucose, glucosamine, galactosamine are found in influenza virus, parainfluenza virus .

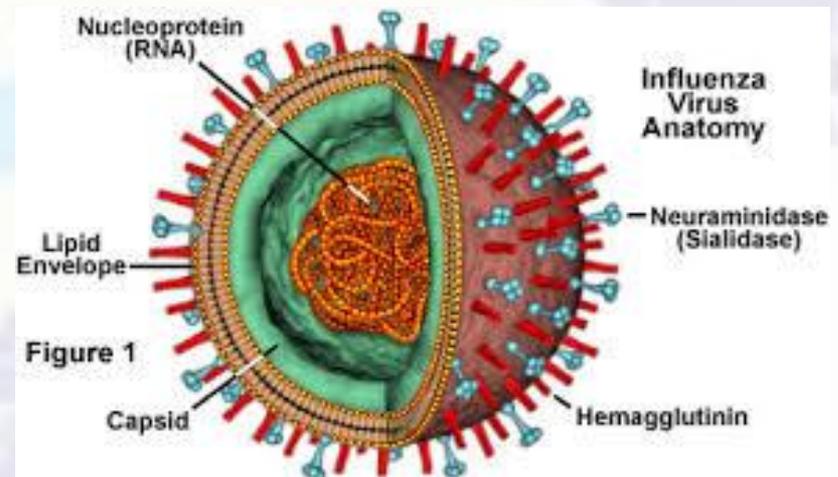


4. Nucleic Acid: Viruses contain either DNA or RNA for their genetic information.

- ❖ Viruses containing DNA are called Deoxyviruses, whereas, having RNA called Riboviruses.
- ❖ In general, all plant viruses have ss-RNA.
- ❖ Animal viruses have either single or (rarely) ds-RNA or ds-DNA.
- ❖ Bacterial viruses contain mostly ds-DNA but can also have ss-DNA or RNA.
- ❖ Insect viruses contain RNA and only a few have DNA.
- ❖ DNA of some bacterial and animal viruses is circular but in others it is like RNA.
- ❖ DNA viruses cause human diseases, such as chickenpox, hepatitis B, and some venereal diseases, like herpes and genital warts.
- ❖ Mutations in RNA viruses occur more frequently than in DNA viruses.
- ❖ This causes them to change and adapt more rapidly to their host.
- ❖ Human diseases caused by RNA viruses include hepatitis C, measles, and rabies.



Hepatitis virus: Deoxyvirus

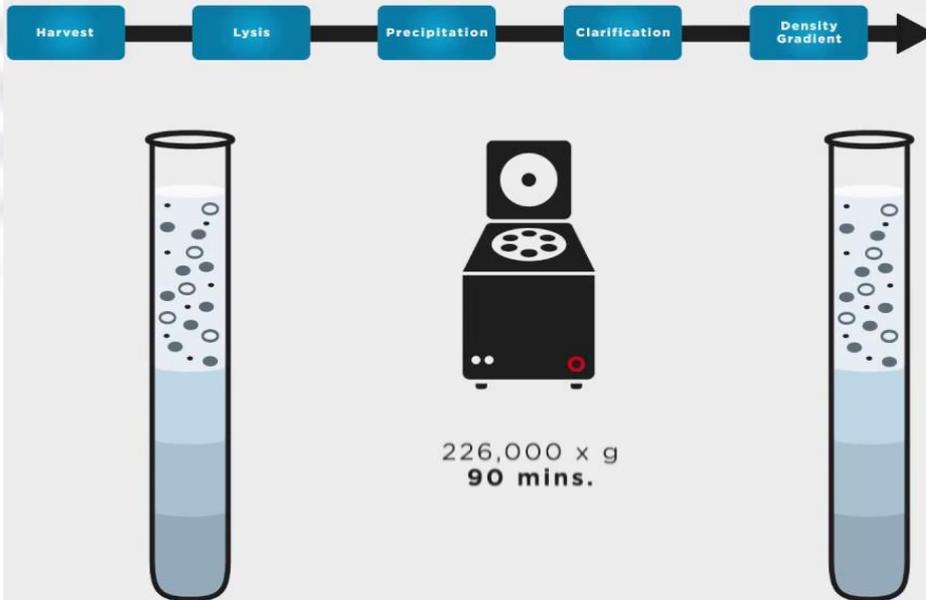


Influenza virus: Ribovirus

ISOLATION AND PURIFICATION OF VIRUSES

❖ The primary aim of virus cultivation are:

- To isolate and identify the virus for clinical specimen
- To detail research on viral structure
- To know the multiplication cycle of virus
- To know the effect on host
- Preparation of vaccine for fatal viral diseases



❖ The following nine main steps involved in the isolation and purification of virus in plants.



1. Virus Propagation in a Suitable Host Plant
2. Selection of Infected Part of the Plant
3. Factors
4. Extraction of Virus Using a Suitable Buffer
5. Infectivity Test
6. Criteria for Purity of Virus
7. Virus Yield
8. Storage of Purified Virus
9. Uses of Purified Virus.

Step 1: Virus Propagation in a Suitable Host Plant

- ❖ A large amount of infected plant material is needed to purify the virus.
- ❖ It can be achieved by inoculation of a water or buffer extract of the infected plant to a number of suitable hosts or to the same healthy host.
- ❖ If a virus cannot be transmitted by mechanical means, it can also be transmitted by grafting or by suitable vector to a healthy host for multiplication.
- ❖ Only those suitable plants as hosts are selected which are free from the chemicals which inhibit infectivity of virus.

Propagation host	Suitable for Virus
<i>Arachis hypogea</i>	PCLV, PCISV
<i>Beta vulgaris</i>	BtMV
<i>Cucumis melo</i>	WMV
<i>Cucumis sativus</i>	CMV, WMV
<i>Nicotiana benthamiana</i>	BYV, SPV ₂
<i>Pisum sativum</i>	MV, PSb
<i>Sorghum bicolor</i>	MMV, SCMV, SCSMV ₁
<i>Vicia faba</i>	CIYVV
<i>Zea mays</i>	MMV, MSV, SCMV
<i>Vigna sinensis</i>	CPMV, TRSV, gemini virus

Step 2: Selection of Infected Part of the Plant

- ❖ Mid rib and petioles contain low concentration of virus.
- ❖ These should be separated before use for virus extraction.
- ❖ Young leaves should be preferred to older leaves.
- ❖ In young leaves inhibitory materials that adsorb or adhere to the virus are frequently low in concentration.
- ❖ Plant parts to be used for virus isolation should not be contaminated by another virus or strain.
- ❖ Roots of pea plant are best to extract clover yellow mosaic virus (C₁YMV).



Step 3: Factors

- ❖ Certain climatic factors like temperature, light intensity and its duration, host nutrition and time after inoculation affect the virus concentration in the host plant.
- ❖ Some virus attain peak concentration in about 12 days after inoculation, rapidly decreased in the next four days and reaches at very low concentration by 48 days.
- ❖ So, the conditions for growth of the infected plants and time of harvest should be standardized for each virus to maximize the virus yield.

Step 4: Extraction of Virus Using a Suitable Buffer

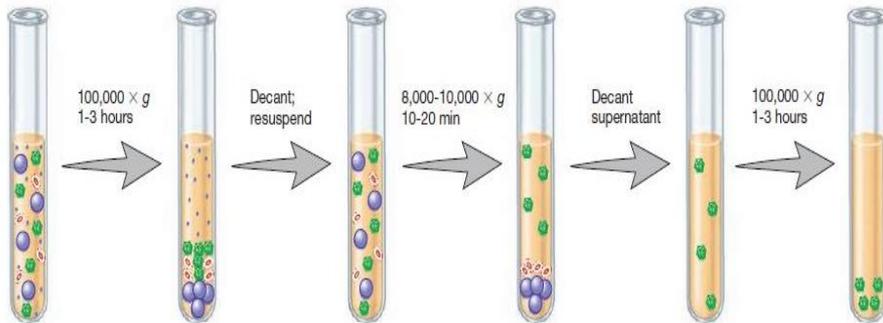
- ❖ Extraction medium must enable extraction of maximum amount of virus from the host plant.
- ❖ Keep the virus in stable, infective, un-aggregation condition and minimize host contaminants.
- ❖ A buffer of suitable pH and molarity with additives to prevent oxidation.
- ❖ Phosphate acetate and borate buffers are commonly used at different pH and molarity.
- ❖ Some virus need a cation like Ca^{2+} or Mg^{2+} to preserve their infectivity, besides the ionic strength of the buffer.
- ❖ Mortar pestle, food blenders, meat mincer and the electrically operated glass mortar and pestle are useful to homogenize the virus infected plant material in a suitable buffer.
- ❖ All extractions of virus are done in cold conditions at 4°C or using salt-ice bath around mortar to prevent oxidation reactions.
- ❖ Cell components (Ribosomes, RuBP carboxylase protein from chloroplast, fragments of lower molecular weight compounds) should be removed in the extraction process, leaving the infective virus in the solution. It can be achieved by the following **methods**:
 - **Heating**
 - **Centrifugation**
 - **Crystallization or Salt precipitation**
 - **Precipitation at Isoelectric Point**
 - **Coacervation**
 - **Gel Filtration**
 - **Density Gradient Electrophoresis**

☐ Heating

- ❖ Heating the extract for a few minutes at 50°-60° helps to coagulate the unwanted materials. It is very good method to extract stable virus TMV.

☐ Centrifugation

- ❖ Centrifugation at different speeds separates virus particles and host components of different densities.
- ❖ Low speed 500-10,000 rpm centrifugation, is used in the initial stages of clarifying the crude infective plant extract to sediment the gross host material.
- ❖ High speed centrifugation at speeds of 30,000 rpm is used at later stages to get relatively pure virus, devoid at most of the host components.



☐ Crystallization

- ❖ Ammonium sulphate to one third saturation of the crude extract is used to precipitate TMV.

☐ Precipitation at Isoelectric Point

- ❖ Virus which are nucleoproteins are precipitated at specific pH. The precipitate is collected by low speed centrifugation and re-suspended in suitable buffer.

☐ Coacervation

- ❖ It separate macromolecules in two liquid layers, one of which is rich in colloid. A virus separates into one of the two layers.

☐ Gel Filtration

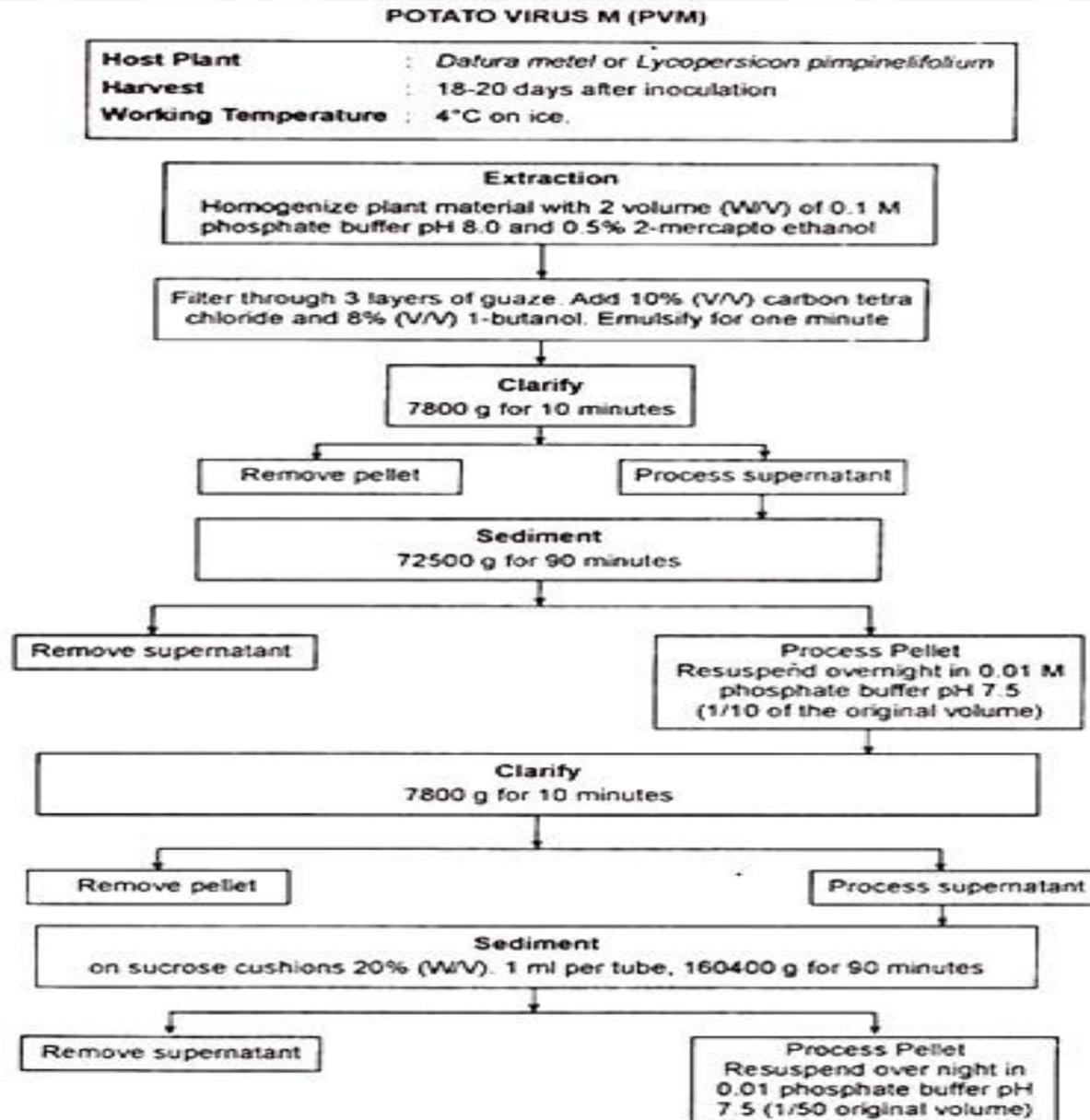
- ❖ Sephadex, agar and agarose remove smaller host components by adsorption.

☐ Density Gradient Electrophoresis

- ❖ Virus components can be separated by layering the preparation on a suitable buffered density gradient formed in a U-tube and applying electrical charge.

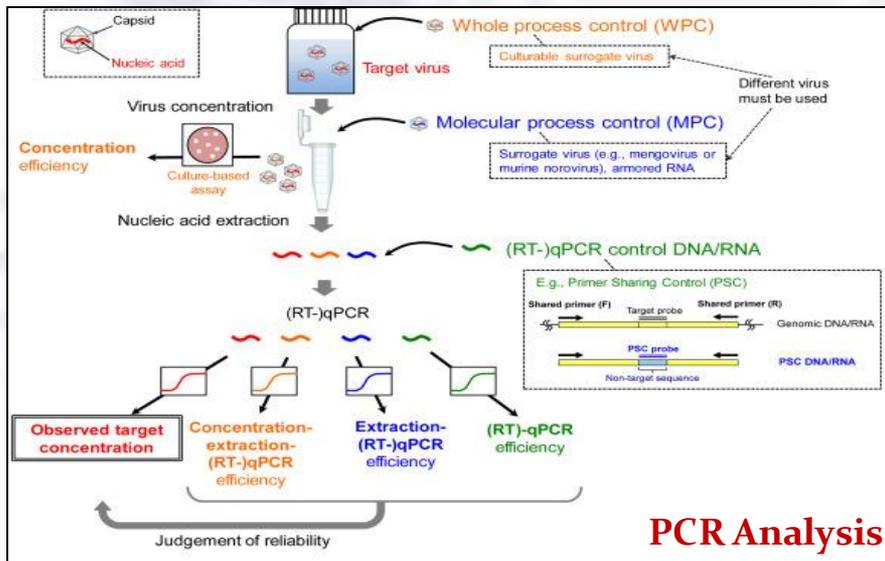
Step 5: Infectivity Test

- ❖ Infectivity of the purified mechanically transmissible virus can be checked by inoculating to a suitable host.



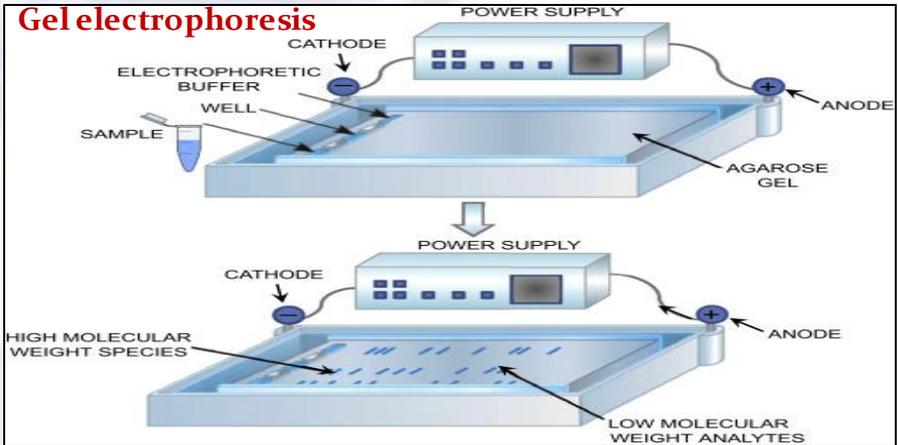
Step 6: Criteria for Purity of Virus

- ❖ Criteria for purity of a virus as a single entity may be by specific characteristic of that virus in a specific test plant, serological tests, UV spectrophotometric absorption data, gel electrophoresis, PCR analysis and molecular characterization.



Step 7: Virus Yield

- ❖ The total weight of purified virus is called virus yield. It varies with the virus and plant used. It ranges from 0.05 ug/g leaf with Barley yellow Dwarf-virus (BYDV) to 2000 µg TMV/g tobacco.

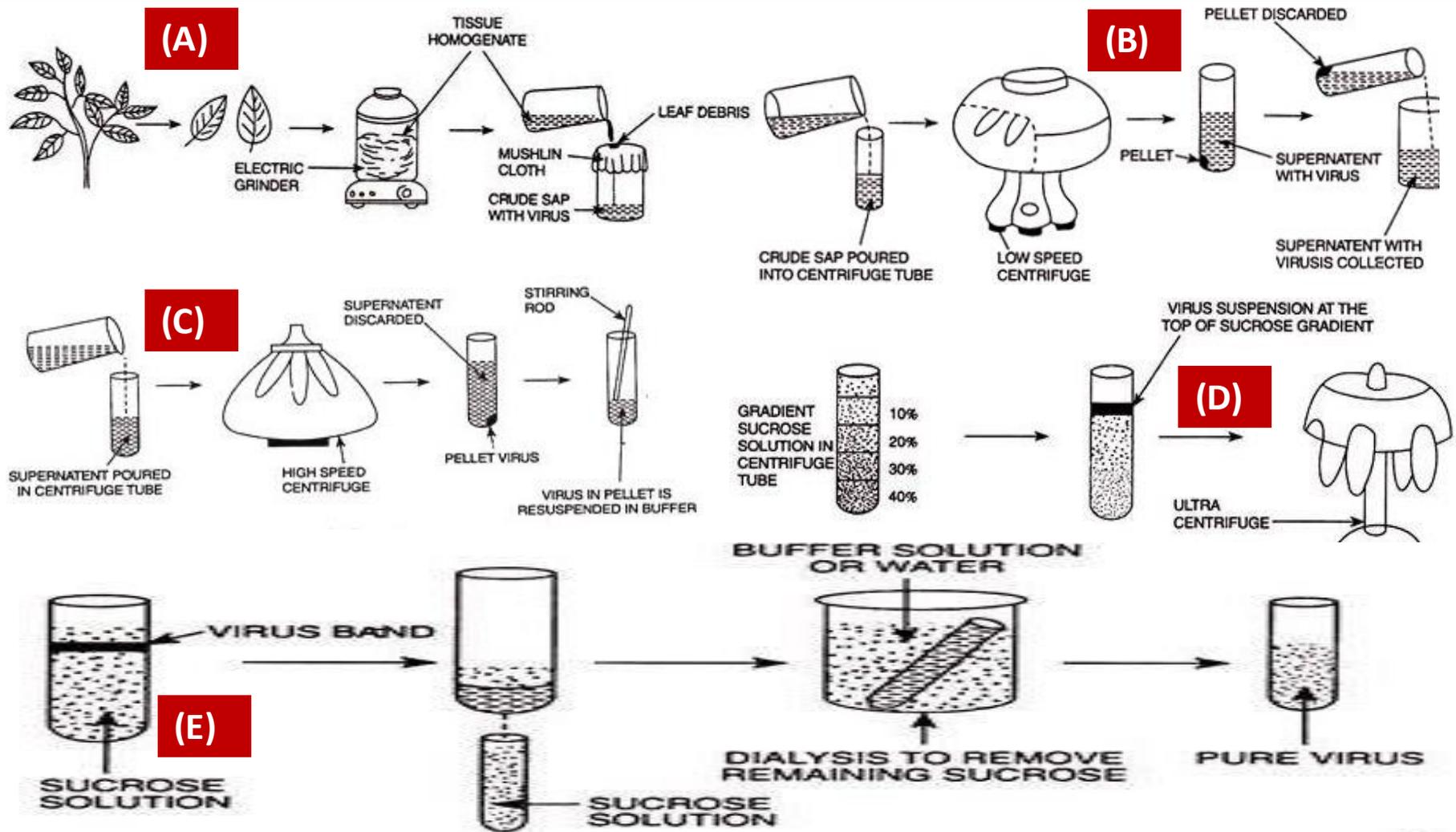


Step 8: Storage of Purified Virus

- ❖ Purified virus is stored in small quantities in vials with equal volume of glycerol and 3 or 4 crystals of thymol or sodium azide (to prevent microbial growth) at 4°C or frozen.

Step 9: Uses of Purified Virus

- ❖ Purified virus free from contaminants is required to determine its structure, biochemical and molecular composition in relation to function and development of probable practicable control or management measures for the disease by various methods, including manipulation of parts of its genome.



Diagrammatic representation for process of purification of virus: (A): Infected leaves homogenized buffer, (B): Crude sap which contains virus is collected and then poured into centrifuge tube in low speed (3000-17000 g). As a result, the crude sap differentiates into supernatant and a pellet. (C): Supernatant is discarded and the pellet of virus is mixed with a buffer and stirred with rod so that it re-suspends in buffer. (D): Virus suspension is placed at top of the top-most layer and centrifuge tube centrifuged in swimming-bucket rotors at high speed ultracentrifuge. (E): When settled, plant virus particles puncture at the bottom of the centrifuge tube. The virus- fraction is placed in cellulose dialysis tubing and sucrose is removed by dialysis in buffer solution or water. Thus, the virus is obtained in pure form .

REPLICATION OF VIRUSES

- ❖ Viral populations do not grow through cell division, because they are acellular.
- ❖ Virus use the machinery and metabolism of a host cell to produce multiple copies of themselves.
- ❖ During the process of **viral replication**, a virus induces a living host cell to synthesize the essential components for the synthesis of new viral particles.
- ❖ The particles are then assembled into the correct structure, and the newly formed virions escape from the cell to infect other cells.
- ❖ The host cell is forced to rapidly produce thousands of identical copies of the original virus.
- ❖ Replication between viruses is varied and depends on the type of genes involved.
- ❖ Most DNA viruses assemble in the nucleus;
- ❖ Most RNA viruses develop solely in cytoplasm.
- ❖ Viral life cycle differs greatly between species, but there are basic stages in their life cycle:
 - Attachment
 - Penetration
 - Uncoating
 - Replication
 - Assembly
 - Release

□ Attachment

- ❖ Attachment is a specific binding between viral capsid proteins and specific receptors on the host cellular surface.
- ❖ This specificity determines the host range and type of host cell of a virus.
- ❖ For example, HIV infects a limited range of human leucocytes.
- ❖ This is because its surface protein, gp120, specifically interacts with the CD4 molecule—a chemokine receptor—which is most commonly found on the surface of CD4+ T-Cells.
- ❖ This mechanism has evolved to favour those viruses that infect only cells in which they are capable of replication.
- ❖ Attachment to the receptor can induce the viral envelope protein to undergo changes that result in the fusion of viral and cellular membranes, or changes of non-enveloped virus surface proteins that allow the virus to enter.

□ Penetration

- ❖ Virions enter the host cell through receptor-mediated endocytosis or membrane fusion. This is often called *viral entry*.
- ❖ The infection of plant and fungal cells is different from that of animal cells.
- ❖ Plants have a rigid cell wall made of cellulose, and fungi one of chitin, so most viruses can get inside these cells only after trauma to the cell wall.
- ❖ However, nearly all plant viruses (such as tobacco mosaic virus) can also move directly from cell to cell, in the form of single-stranded nucleoprotein complexes, through pores called plasmodesmata.
- ❖ Bacteria, like plants, have strong cell walls that a virus must breach to infect the cell. However, since bacterial cell walls are much less thick than plant cell walls due to their much smaller size, some viruses have evolved mechanisms that inject their genome into the bacterial cell across the cell wall, while the viral capsid remains outside.

□ Uncoating

- ❖ In this process viral capsid is removed: This may be by degradation by viral enzymes or host enzymes or by simple dissociation.
- ❖ the end-result is the releasing of the viral genomic nucleic acid.

□ Replication

- ❖ It involves synthesis of viral messenger RNA (mRNA) from "early" genes (with exceptions for positive sense RNA viruses), viral protein synthesis, possible assembly of viral proteins, then viral genome replication mediated by early or regulatory protein expression.
- ❖ This may be followed, for complex viruses with larger genomes, by one or more further rounds of mRNA synthesis: "late" gene expression is, in general, of structural or virion proteins.

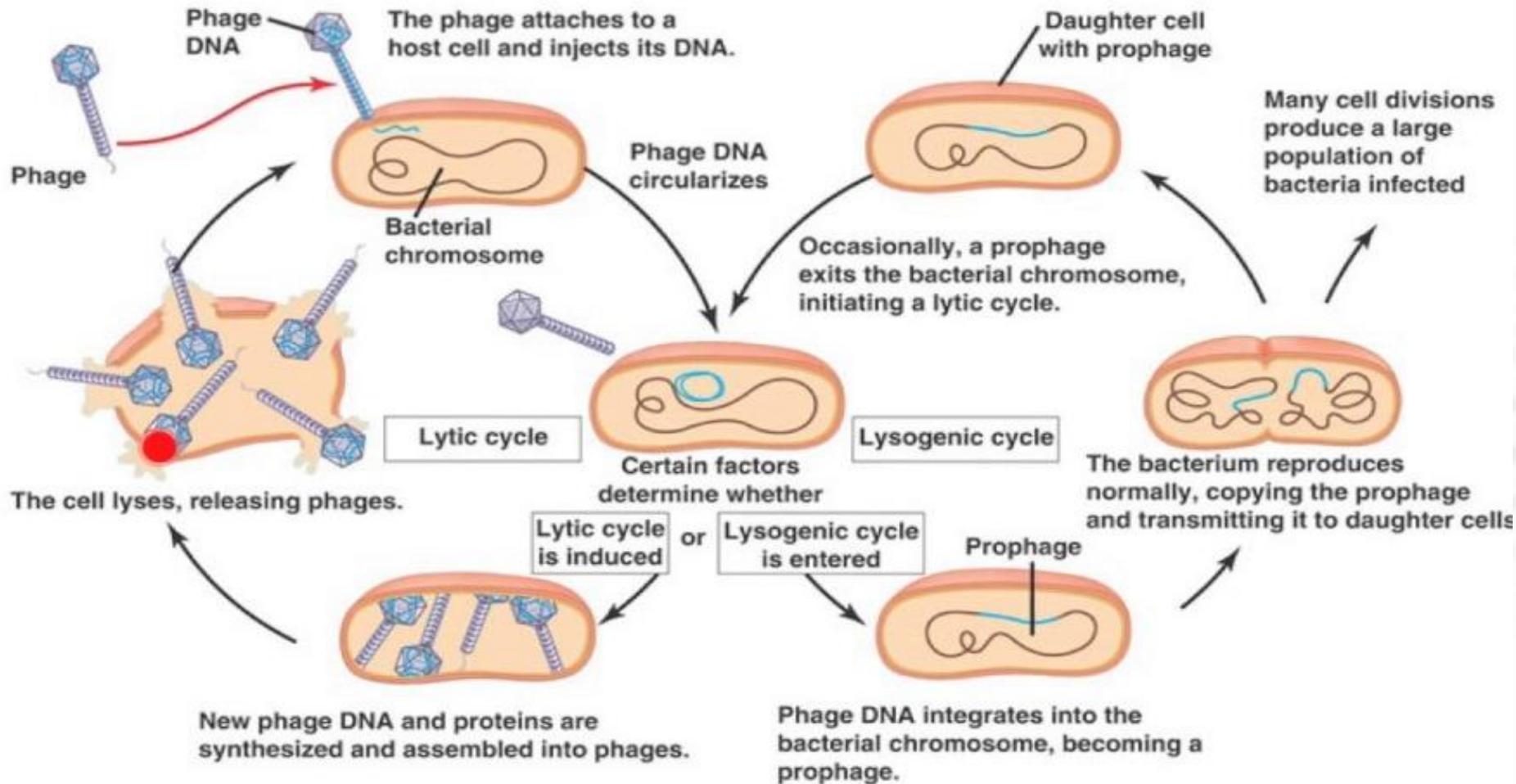
□ Assembly

- ❖ Following the structure-mediated self-assembly of the virus particles, some modification of the proteins often occurs.
- ❖ Viruses such as HIV, modification occurs after the virus has been released from the host cell.

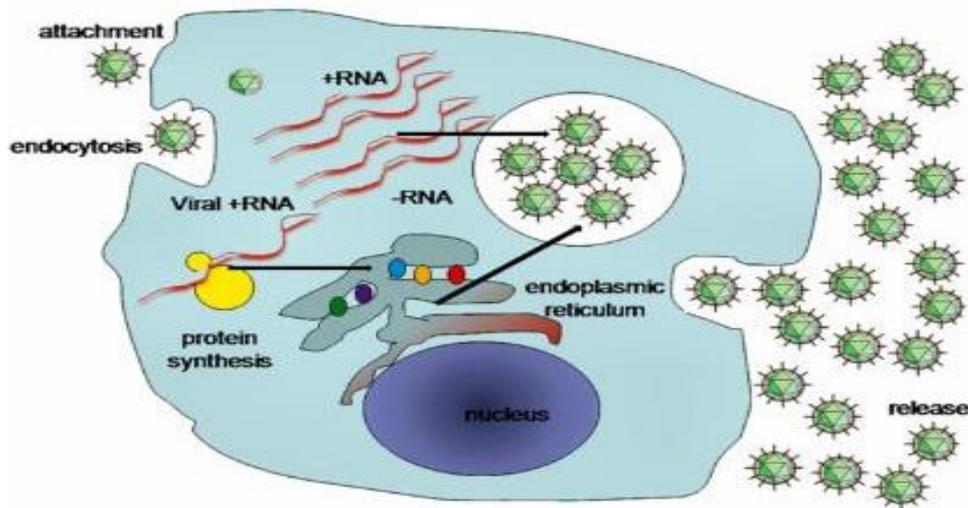
□ Release

- ❖ Viruses can be released from the host cell by lysis, a process that kills the cell by bursting its membrane and cell wall.
- ❖ This is a feature of many bacterial and some animal viruses and called lytic cycle.
- ❖ Some viruses undergo a lysogenic cycle.
- ❖ In lysogenic cycle, viral genome is incorporated by genetic recombination into a specific place in the host's chromosome.
- ❖ The viral genome is then known as a "provirus" or, in the case of bacteriophages a "prophage".
- ❖ Whenever the host divides, the viral genome is also replicated.
- ❖ The viral genome is mostly silent within the host.
- ❖ At some point, the provirus or prophage may give rise to active virus, which may lyse the host cells.
- ❖ Enveloped viruses (e.g., HIV) typically are released from the host cell by budding.
- ❖ During this process the virus acquires its envelope, which is a modified piece of the host's plasma or other, internal membrane.

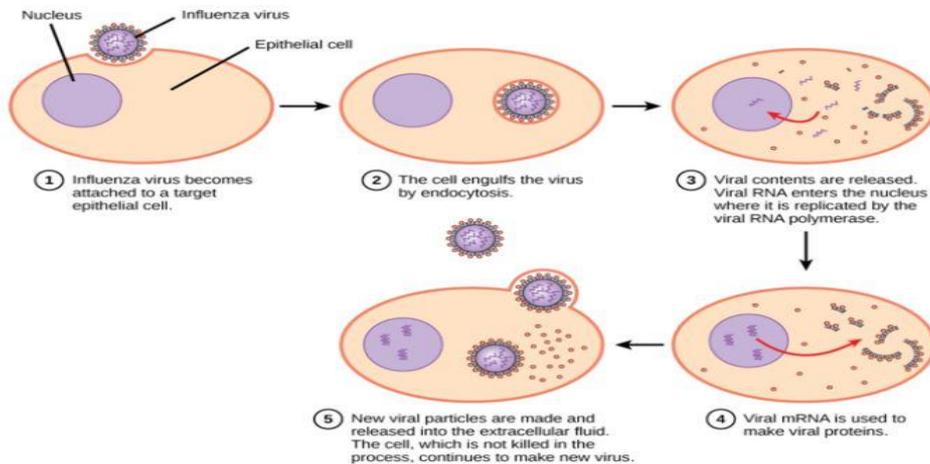
Lytic Vs Lysogenic Mode of Replication of Virus



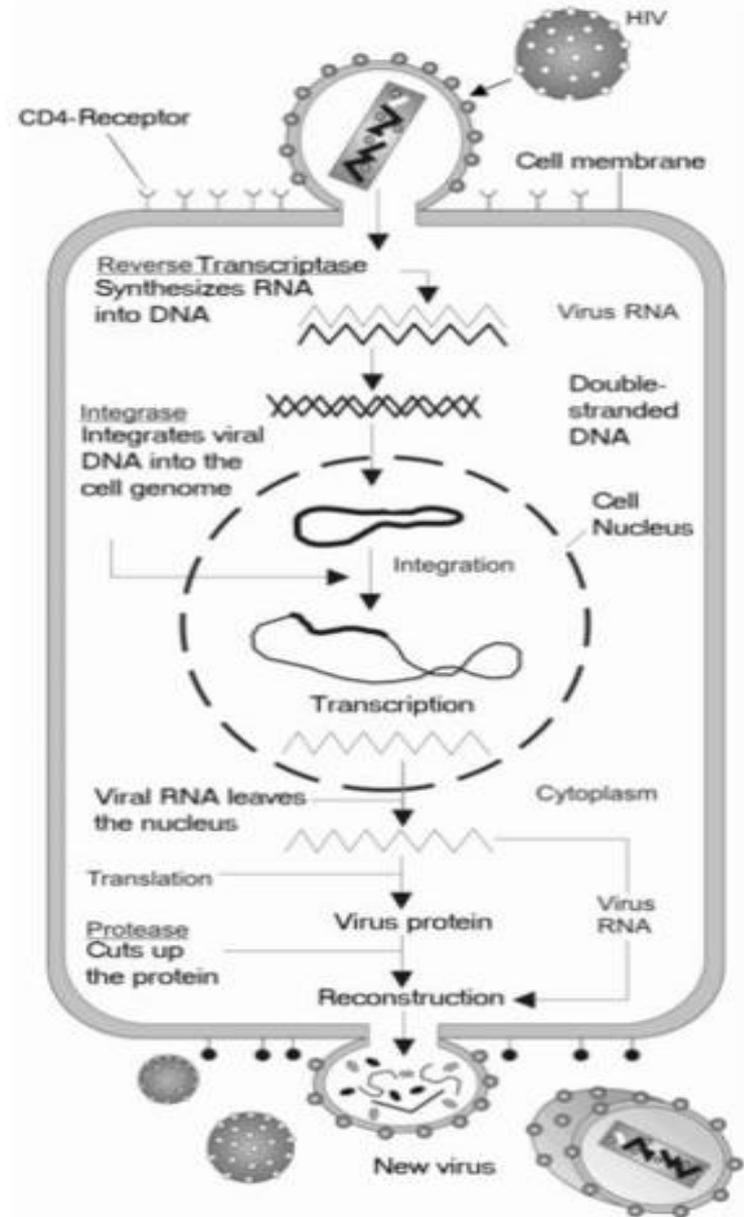
The lytic cycle is often also called the reproductive cycle of a bacteriophage
The virus DNA enters the cell and the virus is repeatedly replicated and the bacterium is lysed
The lysogenic cycle doesn't kill the bacteria
The lysogenic cycle creates many copies of the viral DNA
Lysogenic phages are called temperate phages while lytic phages are virulent



Hepatitis C virus: A simplified diagram of the Hepatitis C virus replication cycle.



Pathway to viral infection: In influenza virus infection, glycoproteins attach to a host epithelial cell. As a result, the virus is engulfed. RNA and proteins are made and assembled into new virions.



Life cycle of HIV

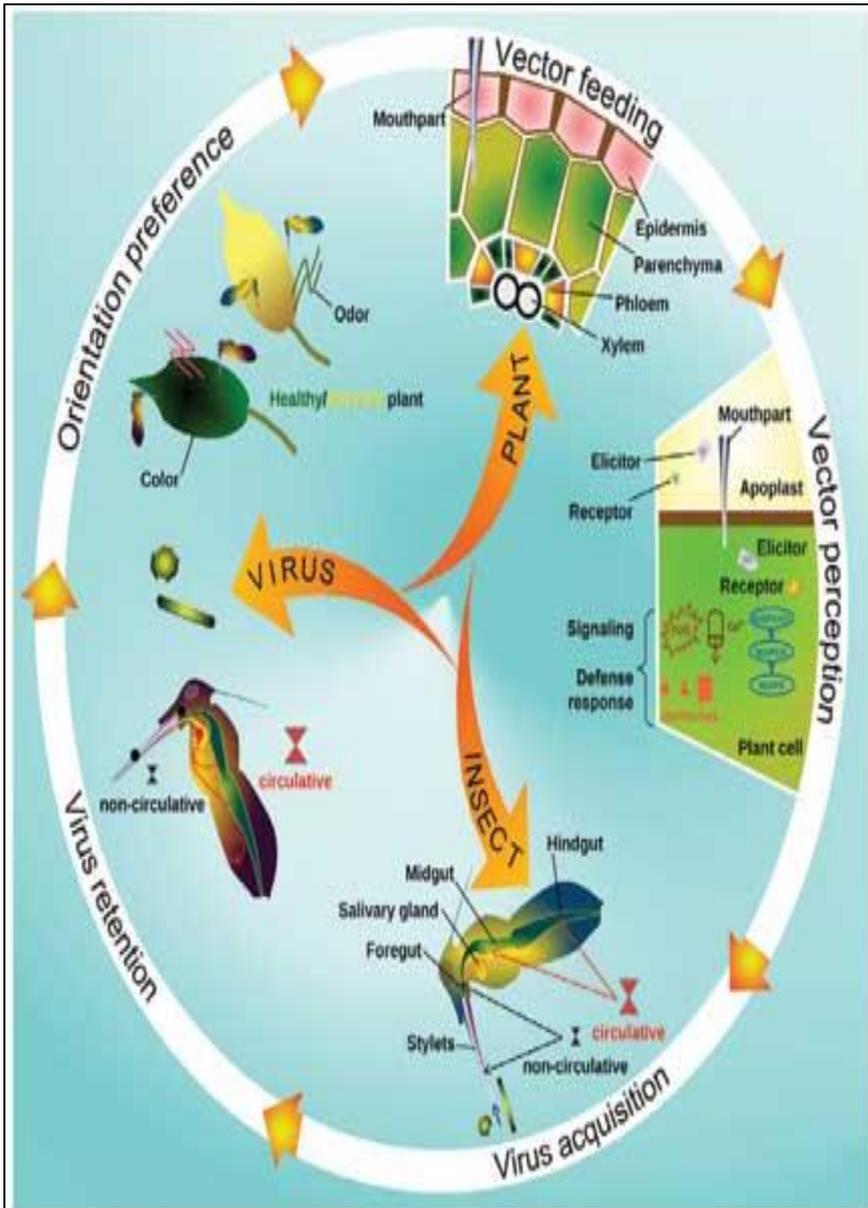
TRANSMISSION OF VIRUSES IN PLANTS

Problems associated with the viral transmission:

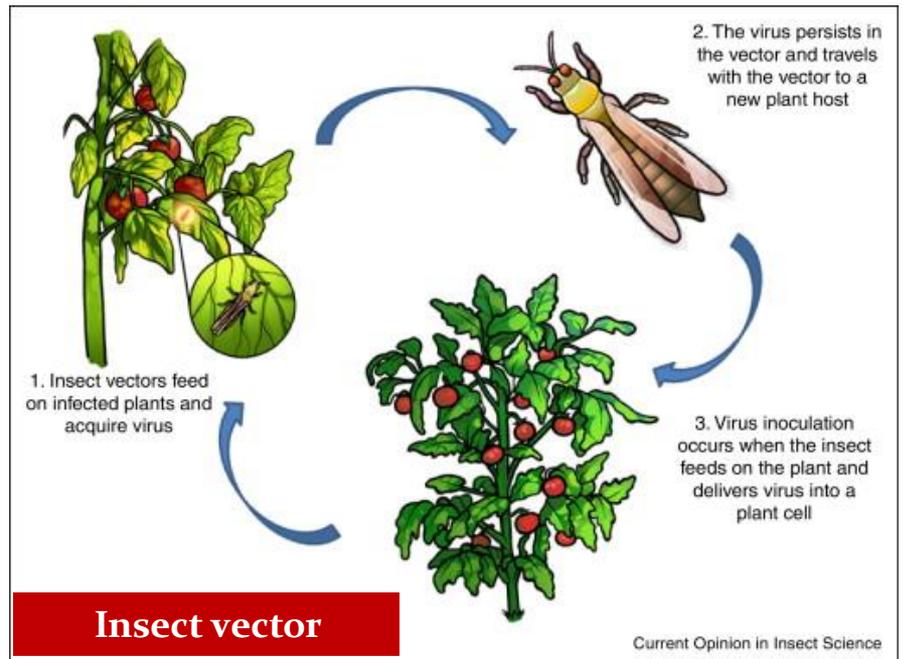
- ❖ Viruses are known to infect both plant cells, animal cells and bacteria.
- ❖ Viruses are obligate intracellular parasites they must develop direct methods of transmission, between hosts, in order to survive.
- ❖ The mobility of animals increases the mechanisms of viral transmission.
- ❖ The plants remain immobile and thus viruses must rely on environmental factors to be transmitted between hosts.

Transmission mode of plant viruses:

- Mechanical transmission by rubbing leaves together, injecting plant extract, by action of animals etc.
- Vegetative and graft transmission through rhizomes, bulbs, corns, tubers etc.
- Pollen transmission; when pollens consisting for viruses fall on stigma of female plants, they germinate and eventually facilitate the virus to infect the ovules of plants.
- Seed transmission; very rare
- Nematode transmission
- Fungal transmission
- Insect vector transmission
- Dodder transmission; dodder are the trailer or climber parasitic plant which grow forming bridge between two plants.

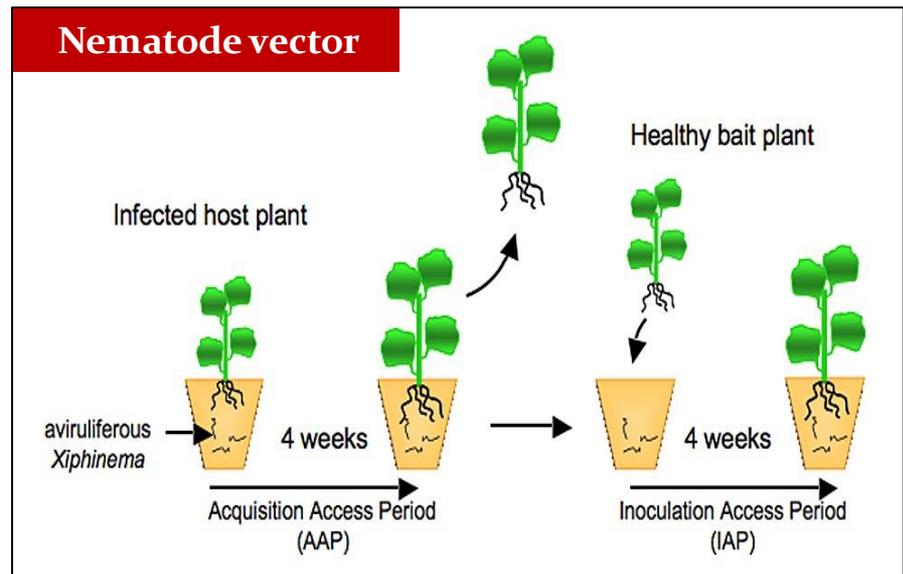


Transmission of virus



Insect vector

Current Opinion in Insect Science



Nematode vector

Natural transmission between plant hosts:

- ❖ In the plants, the cell wall forms a tough barrier between the intracellular components and the extracellular environment, which has to be penetrated.
- ❖ The plant viruses relying on the wind and soil to transmit seeds as well as vectors.
- ❖ Vectors either transmit the virus propagative transmission, which results in an amplification of the virus by replication.
- ❖ Common vectors include Bacteria, Fungi, Nematodes, arthropods and arachnids.
- ❖ Furthermore, human intervention, including grafting and experimental mechanical damage, physically damages the cell wall, contributes to the array of transmission routes.
- ❖ The virus commonly uses these methods to be passed from one host to another.
- ❖ However, the virus is dependent upon physical damage, generated naturally by the wind and feeding of vectors or by human intervention.

Transmission between plant cells:

- ❖ Viral infections often develop into systemic infections as a means of transmission.
- ❖ The virus often infects many tissues, if not the whole plant.
- ❖ There are a variety of methods the virus can use to spread throughout the organism
- ❖ The most common route use the vascular system, known as the xylem and phloem, and the plasmodesmata, which interconnect adjacent cells.
- ❖ The common mechanism involve by different virus is expressing proteins which coat the virus and interact with the structure of the plasmodesmata.
- ❖ The array of proteins expressed by the different viruses may act differently but all achieve a similar goal, passage between adjacent cells.

GENERAL ACCOUNT OF PLANT, ANIMAL AND HUMAN VIRAL DISEASE

What Is Disease ????

The definition of a disease is an illness or sickness with specific, well-defined symptoms that affects a person, plant or animal.

Or

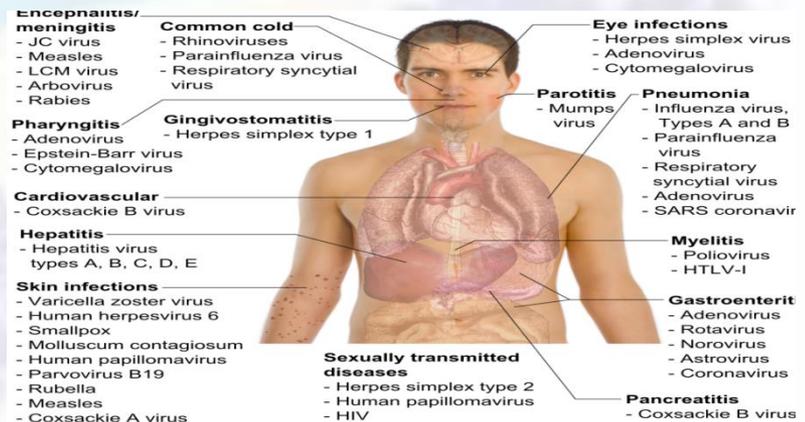
A condition of the living animal or plant body or of one of its parts that impairs normal functioning and is typically manifested by distinguishing signs and symptoms

Or

A disorder of structure or function in a human, animal, or plant, especially one that produces specific symptoms or that affects a specific location and is not simply a direct result of physical injury.

Or

A disease is a particular abnormal condition that negatively affects the structure or function of all or part of an organism, and that is not due to any immediate external injury. Diseases are often known to be medical conditions that are associated with specific symptoms and signs.



Types of Causes of Diseases

- ❖ **Airborne:** Disease that is caused by pathogens and transmitted through the air.
- ❖ **Food borne:** Illness resulting from the consumption of food contaminated with
 - *Bacteria*
 - *Toxins*
 - *Viruses*
 - *Prions*
 - *Parasites*
- ❖ **Infectious:** It is known as transmissible diseases or communicable diseases, comprise clinically evident illness (i.e., characteristic medical signs or symptoms of disease) resulting from the infection, presence and growth of pathogenic biological agents in an individual host organism. Included in this category are:
 - *Contagious diseases*—commonly spreads from one person to another. Eg., influenza
 - *Communicable diseases*—a disease that can spread from one person to another, but does not necessarily spread through everyday contact.
- ❖ **Lifestyle:** A lifestyle disease is any disease that appears to increase in frequency as countries become more industrialized and people live longer, especially if the risk factors include behavioral choices like a sedentary lifestyle or a diet high in unhealthful foods such as refined carbohydrates, trans fats, or alcoholic beverages.
- ❖ **Non-communicable:** A non-communicable disease is a medical condition or disease that is non-transmissible. Non-communicable diseases cannot be spread directly from one person to another. Heart disease and cancer are examples of non-communicable diseases in humans.

Plant Virus Diseases

- ❖ Plant viruses are viruses that affect plants and obligate intracellular parasites.
- ❖ Plant viruses can be pathogenic to higher plants.
- ❖ Most plant viruses are rod-shaped, with protein discs forming a tube surrounding the viral genome; isometric particles are another common structure.
- ❖ They rarely have an envelope.
- ❖ The great majority have an RNA genome, which is usually small and single stranded (ss), but some viruses have double-stranded (ds) RNA, ssDNA or dsDNA genomes.
- ❖ Tobacco mosaic virus (TMV), the first virus to be discovered.
- ❖ Plant viruses are grouped into 73 genera and 49 families. However, these figures relate only to cultivated plants, which represent only a tiny fraction of the total number of plant species.
- ❖ Viruses in wild plants have been relatively little studied.
- ❖ To transmit from one plant to another and from one plant cell to another, plant viruses must use strategies that are usually different from animal viruses.
- ❖ Plants do not move, and so plant-to-plant transmission usually involves vectors (such as insects).
- ❖ Plant cells are surrounded by solid cell walls, therefore transport through plasmodesmata is the preferred path for virions to move between plant cells.
- ❖ Plants have specialized mechanisms for transporting mRNAs through plasmodesmata, and these mechanisms are thought to be used by RNA viruses to spread from one cell to another.
- ❖ Plant defenses against viral infection include, among other measures, the use of siRNA in response to dsRNA.
- ❖ Most plant viruses encode a protein to suppress this response.
- ❖ Plants also reduce transport through plasmodesmata in response to injury.

Structure of Plant Viruses

- ❖ Plant viruses are extremely small and can only be observed under an electron microscope.
- ❖ The structure of a virus is given by its coat of proteins, which surround the viral genome.
- ❖ Assembly of viral particles takes place spontaneously.
- ❖ Over 50% of known plant viruses are rod-shaped (flexuous or rigid).
- ❖ They are usually between 300–500 nm lengths with a diameter of 15–20 nm.
- ❖ Protein subunits can be placed around the circumference of a circle to form a disc.
- ❖ In the presence of the viral genome, the discs are stacked, then a tube is created with room for the nucleic acid genome in the middle.
- ❖ The second most common structure amongst plant viruses are isometric particles.
- ❖ They are 25–50 nm in diameter. In cases when there is only a single coat protein, the basic structure consists of $60T$ subunits, where T is an integer.
- ❖ Some viruses may have 2 coat proteins that associate to form an icosahedral shaped particle.
- ❖ There are three genera of *Geminiviridae* that consist of particles that are like two isometric particles stuck together.
- ❖ A very small number of plant viruses have, in addition to their coat proteins, a lipid envelope.
- ❖ This is derived from the plant cell membrane as the virus particle buds off from the cell.

Transmission of Plant Viruses

❖ **Through sap:** Viruses can be spread by direct transfer of sap (E.g., TMV, potato viruses and cucumber mosaic viruses)by contact of a wounded plant with a healthy one. Such contact may occur:

1. By damage caused by tools
2. By hands
3. By naturally
4. By an animal feeding on the plant.

❖ **Insect:** Plant viruses transmitted by a vector, most often insects (E.g., leafhoppers).Class Rhabdoviridae, has been proposed to actually be insect viruses that have evolved to replicate in plants. The chosen insect vector of a plant virus will often be the determining factor in that virus's host range. Depending on the way, plant viruses are classified as:

1. **Non-persistent:** Viruses become attached to the distal tip of the stylet of the insect and on the next plant it feeds on, it inoculates it with the virus.
2. **Semi-persistent:** Virus entering the foregut of the insect.
3. **Persistent:** Those viruses that manage to pass through the gut into the haemolymph and then to the salivary glands. There are two sub-classes of persistent viruses:
 - a. *Propagative:* Propagative viruses are able to replicate in both the plant and the insect (and may have originally been insect viruses).
 - b. *Circulative:* Must escape the insect gut and spread to neighboring organs to reach the salivary glands for transmission.

- ❖ **Nematodes:** Soil-borne nematodes also transmit viruses. They acquire and transmit them by feeding on infected roots. The virions attach to the stylet (feeding organ) or to the gut when they feed on an infected plant and can then detach during later feeding to infect other plants. E.g., Tobacco Ringspot Virus and Tobacco Rattle Virus.
- ❖ **Plasmodiophorids:** A number of virus genera are transmitted, both persistently and non-persistently, by soil borne zoosporic protozoa. These protozoa are not phytopathogenic themselves, but parasitic. Transmission of the virus takes place when they become associated with the plant roots. Examples *Polymyxa graminis*, and *Polymyxa betae* which transmits Beet necrotic yellow vein virus. Plasmodiophorids also create wounds in the plant's root through which other viruses can enter.
- ❖ **Seed and pollen borne viruses:** In this mode of transmission, the seed is infected in the generative cells and the virus is maintained in the germ cells and sometimes, but less often, in the seed coat. E.g., Bean common mosaic virus. When the growth and development of plants is delayed because of situations like unfavorable weather, there is an increase in the amount of virus infections in seeds. Little is known about the mechanisms involved in the transmission of plant viruses via seeds, although it is known that it is environmentally influenced and that seed transmission occurs because of a direct invasion of the embryo
 - via the ovule
 - by an indirect route with an attack on the embryo
 - by infected gametes.
- ❖ **Direct plant-to-human transmission:** The virus may be transmitted through the infected human. This is a very rare and highly unlikely event as, to enter a cell and replicate, a virus must bind to a receptor on its surface, and a plant virus would be highly unlikely to recognize a receptor on a human cell. E.g., Pepper Mild Mottle Virus (PMMoV) may have moved on to infect humans.

Pathogenesis

- ❖ One of the important characteristics of pathogenic organisms, in terms of their ability to infect, is virulence.
- ❖ Pathogenesis is the stage of disease in which the pathogen is in intimate association with living host tissue. Three fairly distinct stages are involved:

1. Inoculation: transfer of the pathogen to the infection court, or area in which invasion of the plant occurs (the infection court may be the unbroken plant surface, a variety of wounds, or natural openings) e.g.,

- Stomata: microscopic pores in leaf surfaces
- Hydathodes: stomata-like openings that secrete water
- Lenticels: small openings in tree bark



Stomata



Hydathods



Lenticels

2. Incubation: the period of time between the arrival of the pathogen in the infection court and the appearance of symptoms.

3. Infection: the appearance of disease symptoms accompanied by the establishment and spread of the pathogen.

Control Measurements of Viral Diseases of Plants

❖ After a plant is infected with a virus/viroid, little can be done to restore its health.

1. *Genetic Host Resistance*

- Control is accomplished by several methods, such as growing resistant species and varieties of plants or obtaining virus-free seed, cuttings, or plants as a result of indexing and certification programs.

2. *Control the vectors*

- Controlling insect vectors by spraying plants with contact insecticides or fumigating soil to kill insects, nematodes, and other possible vectors.
- Growing valuable plants under fine cheesecloth or wire screening that excludes vectors.

3. *Cultural Practices*

- Separation of new from virus-infected plantings of the same or closely related species.
- For cutting, grafting or propagating seedlings vegetatively, use cleaner or sanitized tools and equipment, and disposable overcoat; wash hands frequently.
- Rotations to non-host crops.
- Geographic isolation of production facilities may also help avoid losses.
- The isolation of newly received plant material prior to its introduction into the rest of a production system can also minimize the unintentional introduction of pathogens.
- Both dry and wet heat treatments are based on the sensitivity of certain viruses to high temperatures.

4. *Chemical Applications and Biological Control*

- There are no chemical sprays or biological control approaches to eradicate viruses,
- Although insecticides and biocontrol products can be used to control insect vectors.

5. *Government Regulatory Measures*

- Management of insect vector populations in the field can be difficult to impossible unless coordinated on a regional basis but may be highly effective in closed production systems such as greenhouses or interiorscapes.

❑ *Cultural Practices of some specific plant species*

- ✓ Infected peach, apple, and rose budwood stock and carnations have been grown for weeks or months at temperatures about 37 to 38 °C (99 to 100 °F) to free new growth from viruses.
- ✓ Soaking some woody plant parts or virus-infected sugarcane shoots in hot water at about 50°C (120 °F) for short periods also is effective.
- ✓ Both dry and wet heat treatments are based on the sensitivity of certain viruses to high temperatures.
- ✓ Rapidly growing dahlia and chrysanthemum sprouts outgrow viruses so that stem tips can be used to propagate healthy plants.
- ✓ With certain carnations, chrysanthemums, and potatoes, a few cells from the growing tip have been grown under sterile conditions in tissue culture; from these, whole plants have been developed free from viruses.

Some Common Viral Diseases of Plants

Disease	Causative Agent	Hosts	Symptoms and Signs
Tobacco mosaic	Tobacco mosaic virus (TMV)	Tobacco, tomato, and hundreds of other vegetables and weeds	Mottled appearance of leaves (mosaic pattern); dwarfing
Cucumber mosaic	Cucumber mosaic virus (CMV)	Cucumber, bean, tobacco, and other plants (wide range of hosts)	The plant turn pale and bumpy. Leaves turn mosaic and wrinkled. Growth stunted. Fruits became oddly shaped and appear gray, and bitter.
Barley yellow dwarf	Barley yellow dwarf virus (BYDV)	Barley, oats, rye, wheat; also pasture grasses and weeds	Yellowing and dwarfing of leaves; stunting of plants
Tomato spotted wilt	Tomato spotted wilt virus (TSWV)	Tomato, pepper, pineapple, peanut, and many other plants	Leaves show concentric, necrotic rings; necrotic region yellow, then turning red-brown
Prunus necrotic ring spot	Prunus necrotic ring spot virus (PNRV)	Stone fruits—e.g., cherry, almond, peach, apricot, plum, and others	Delayed foliation; leaves on infected branches show light green spots and dark rings, then become necrotic and fall off
Potato spindle tuber	Potato spindle tuber viroid (PSTV)	Potato and tomato	Stunted growth; tubers are spindle-shaped and smaller than healthy tubers
Citrus exocortis	Citrus exocortis viroid (CEV)	Orange, lemon, lime, and other citrus plants	Infected trees show vertical splits in bark, thin strips of partially loosened bark, and a cracked, scaly appearance
Yellow leaf curl of Tomato	Tomato yellow leaf curl virus (TYLCV)	<i>Bemisia tabaci</i>	Plants become severely stunted and shoots become erect. Leaflets are reduced in size, curl upwards, become distorted, and have prominent yellowing along margins and/or interveinal regions. Flowers wither

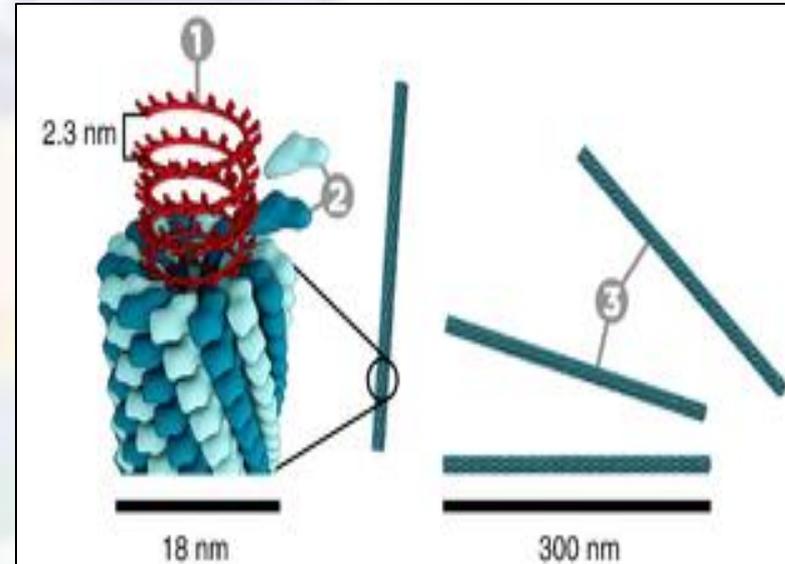


Symptoms of some common diseases: (A) Tobacco mosaic, (B) Cucumber mosaic (C) Barley yellow dwarf, (D) Tomato spotted wilt, (E) Prunus necrotic ring spot (F) Potato spindle tuber (G) Citrus exocortis (H) Yellow leaf curl of Tomato

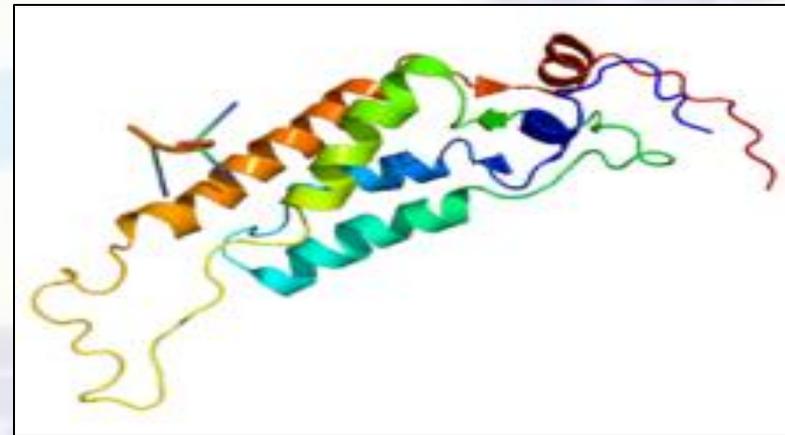
Tobacco Mosaic Disease

❖ Causal Agent: *Tobacco mosaic virus* (TMV)

- It is a single stranded RNA virus in the genus *Tobamovirus*.
- It is the first pathogen identified as a virus.
- *Tobacco mosaic virus* has a rod-like appearance.
- Its capsid is made from 2130 molecules of coat protein and one molecule of genomic single strand RNA, 6400 bases long.
- The coat protein self-assembles into the rod-like helical structure (16.3 proteins per helix turn) around the RNA, which forms a hairpin loop structure.
- The protein monomer consists of 158 amino acids which are assembled into four main alpha-helices, which are joined by a prominent loop proximal to the axis of the virion.
- Virions are ~300 nm in length and ~18 nm in diameter.
- Inside the capsid helix, near the core, is the coiled RNA molecule, which is made up of $6,395 \pm 10$ nucleotides.



Schematic model of TMV: 1. nucleic acid (RNA), 2. capsomer protein (protomer), 3. capsid



A monomeric unit of the *Tobacco mosaic virus* coat protein.

❖ **Transmission Vector**

- Aphids
- Grasshopper

❖ **Physicochemical Properties**

- TMV is a thermostable virus. It can withstand up to 50 °C (120 degree Fahrenheit) for 30 minutes.
- TMV has an index of refraction of about 1.57.

❖ **Disease Cycle**

- TMV can survive in contaminated tobacco products for many years.
- With the direct contact with host plants through its vectors, TMV will go through the infection process and then the replication process.

❖ **Infection and Transmission**

- TMV after its multiplication, it enters the neighbouring cells through plasmodesmata.
- The infection spreads by direct contact to the neighbouring cells.
- For its smooth entry, TMV produces a 30 kDa movement protein called P₃₀ which enlarges the plasmodesmata.
- TMV most likely moves from cell-to-cell as a complex of the RNA, P₃₀, and replicate proteins.
- It can also spread through phloem for longer distance movement within the plant.
- TMV can be transmitted from one plant to another by direct contact.
- Although TMV does not have defined transmission vectors, the virus can be easily transmitted from the infected hosts to the healthy plants, by human handling.

❖ Host and Symptoms

- TMV has a very wide host range and has different effects depending on the host being infected.
- The first symptom of this virus disease is a light green coloration between the veins of young leaves.
- This is followed quickly by the development of a "mosaic" or mottled pattern of light and dark green areas in the leaves.
- Infected plant leaves display small localized random wrinkles.
- These symptoms develop quickly and are more pronounced on younger leaves.
- Its infection does not result in plant death, but if infection occurs early in the season, plants are stunted.
- Lower leaves are subjected to "mosaic burn" especially during periods of hot and dry weather.
- In these cases, large dead areas develop in the leaves.
- This constitutes one of the most destructive phases of *Tobacco mosaic virus* infection.
- Infected leaves may be crinkled, puckered, or elongated.



❖ Environment for disease spread

- TMV is known as one of the most stable viruses. It has a very wide survival range.
- As long as the surrounding temperature remains below approximately 40 degrees Celsius, TMV can sustain its stable form.
- Greenhouses and botanical gardens would provide the most favorable condition for TMV to spread out, due to the high population density of possible hosts and the constant temperature throughout the year.

❖ Treatment and management

- One of the common control methods for TMV is sanitation, which includes removing infected plants and washing hands in between each planting.
- Crop rotation should also be employed to avoid infected soil/seed beds for at least two years.
- Resistant strains against TMV may also be advised.
- The cross protection method can be administered, where the stronger strain of TMV infection is inhibited by infecting the host plant with mild strain of TMV, similar to the effect of a vaccine.
- Application of genetic engineering on a host plant genome has been developed to allow the host plant to produce the TMV coat protein within their cells.
- It was hypothesized that the TMV genome will be re-coated rapidly upon entering the host cell, thus it prevents the initiation of TMV replication.
- Later it was found that the mechanism that protects the host from viral genome insertion is through gene silencing (Agrios , 2005).

Animal and Human Virus Diseases

- ❖ Animal viruses are viruses that affect animals. Animal viruses may be divided into two class:
 1. Vertebrates
 2. Invertebrates

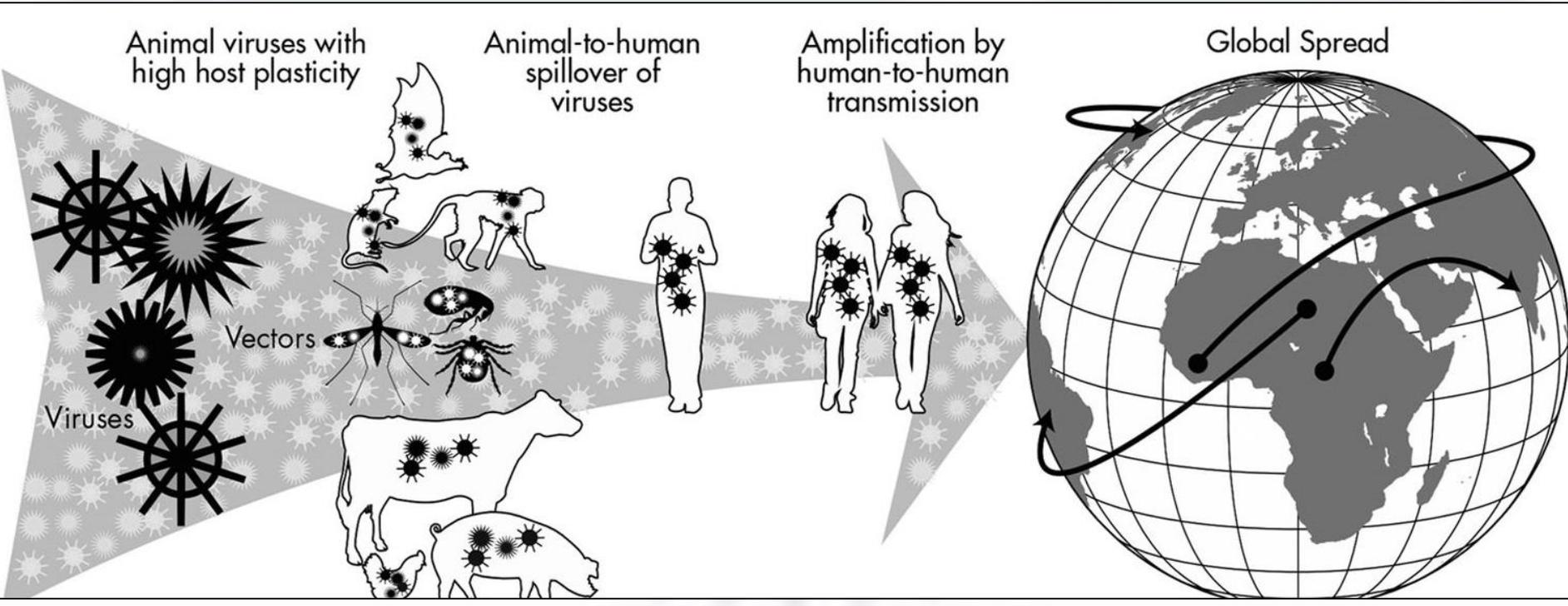
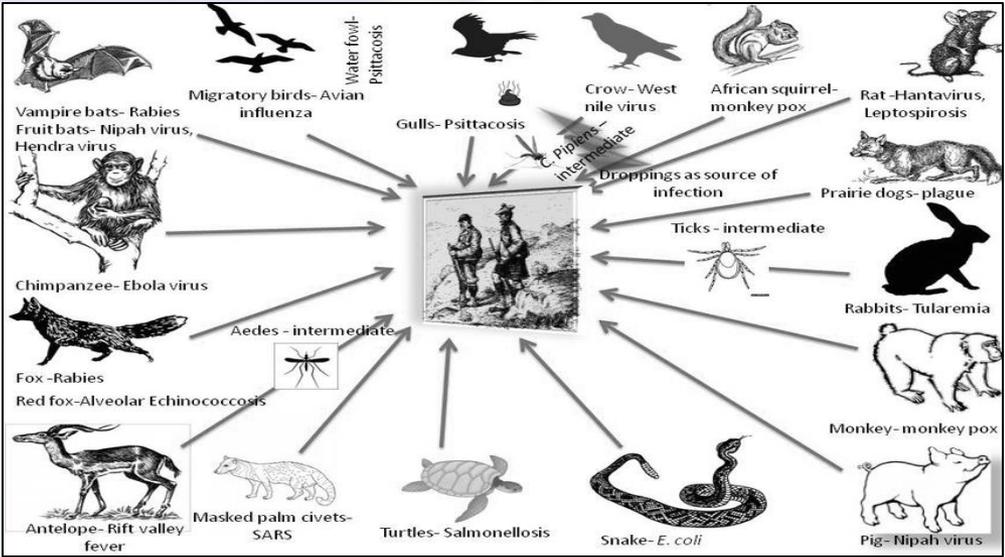
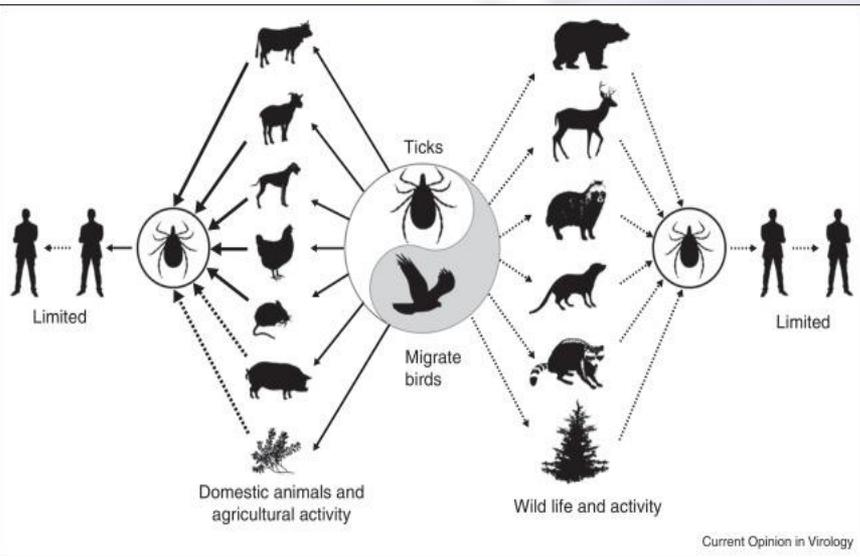
❑ Viral Diseases of Vertebrates

- ❖ The viruses of vertebrates are causes infections of humans and animals.
- ❖ The two fields of study are called clinical virology and veterinary virology, respectively.
- ❖ Different viruses can infect all the organs and tissues of the body and the outcomes range from mild or no symptoms, to life-threatening diseases.
- ❖ Humans cannot be infected by plant or insect viruses, but they are susceptible to infections with viruses from other vertebrates, called viral zoonoses or zoonotic infections. Examples include, rabies, yellow fever and pappataci fever.
- ❖ Viruses are important pathogens of livestock and cause diseases such as foot-and-mouth disease and bluetongue.
- ❖ Jersey and Guernsey breeds of cattle are particularly susceptible to pox viruses.
- ❖ Animals pups such as cats, dogs, and horses, if not vaccinated, can catch serious fatal viral infections Canine parvovirus 2, caused by a small DNA virus.
- ❖ Many other viruses, including caliciviruses, herpesviruses, adenoviruses and parvoviruses, circulate in marine mammal populations.
- ❖ Fish too have their viruses. They are particularly prone to infections with rhabdoviruses.
- ❖ At least nine types of rhabdovirus cause economically important diseases in species including salmon, pike, perch, sea bass, carp and cod.

❑ Viral Diseases of Invertebrates

- ❖ The Arthropods is the largest group of animals and has shown to be a major reservoir of different viruses, both insect-specific viruses (ISV) and viruses that can infect both vertebrates and invertebrates, more known as arthropod-borne viruses (arboviruses).
- ❖ Insect-specific viruses are, as the name reveals, characterised by their incapacity to infect vertebrates.
- ❖ This can be assessed through, viral inoculation of mammalian, avian, or amphibian cell lines.
- ❖ The first (ISV) was discovered over 40 years ago by Stollar and Thomas. It was isolated from an *Aedes aegypti* cell culture.
- ❖ Invertebrates do not produce antibodies by the lymphocyte-based adaptive immune system that is central to vertebrate immunity, but they are capable of effective immune responses.
- ❖ The honey bee (*Apis mellifera*) is susceptible to many viral infections, and often suffer infestations of varroa mites, which are vectors for deformed wing virus.
- ❖ This virus has become one of the most widely distributed and contagious insect viruses on the planet.
- ❖ Baculoviruses are among the best studied of the invertebrate viruses. They infect and kill several species of agricultural pests, and as natural insecticides, they have been used to control insect populations in Brazil and Paraguay such as the velvet bean caterpillar (*Anticarsia gemmatalis*), a pest of soy beans.
- ❖ A baculovirus of the gypsy moth (*Lymantria dispar*) makes their caterpillars climb to the tops of trees where they die. In doing so, they release a shower of millions of progeny viruses that go on to infect more caterpillars.

Transmission of Animal and Human Virus Diseases



Some Common Viral Diseases of Human and animals

Virus	Host	Transmission	Disease	
Adeno-associated virus	Dependovirus, Parvoviridae	Human, vertebrates	Respiratory	None
Australian bat lyssavirus	Lyssavirus, Rhabdoviridae	Human, bats	Zoonosis, animal bite	Fatal encephalitis
Banna virus	Seadornavirus, Reoviridae	Human, cattle, pig, mosquitoes	Zoonosis, arthropod bite	Encephalitis
Barmah forest virus	Alphavirus, Togaviridae	Human, marsupials, mosquitoes	Zoonosis, arthropod bite	Fever, joint pain
Bunyamwera virus	Orthobunyavirus, Bunyaviridae	Human, mosquitoes	Zoonosis, arthropod bite	Encephalitis
Bunyavirus La Crosse	Orthobunyavirus, Bunyaviridae	Human, deer, mosquitoes, tamias	Zoonosis, arthropod bite	Encephalitis
Bunyavirus snowshoe hare	Orthobunyavirus, Bunyaviridae	Human, rodents, mosquitoes	Zoonosis, arthropod bite	Encephalitis
Cercopithecine herpesvirus	Lymphocryptovirus, Herpesviridae	Human, monkeys	Zoonosis, animal bite	Encephalitis
Chandipura virus	Vesiculovirus, Rhabdoviridae	Human, sandflies	Zoonosis, arthropod bite	Encephalitis
Chikungunya virus	Alphavirus, Togaviridae	Human, monkeys, mosquitoes	Zoonosis, arthropod bite	Fever, joint pain
Cowpox virus	Orthopoxvirus, Poxviridae	Human, mammals	Zoonosis, contact	None
Coxsackievirus	Enterovirus, Picornaviridae	Human	Fecal-oral	Meningitis, myocarditis, paralysis
Dengue virus	Flavivirus, Flaviviridae	Human, mosquitoes	Zoonosis, arthropod bite	Hemorrhagic fever

Dhori virus	Thogotovirus, Orthomyxoviridae	Human, ticks	Zoonosis, arthropod bite	Fever, encephalitis
Duvenhage virus	Lyssavirus, Rhabdoviridae	Human, mammals	Zoonosis, animal bite	Fatal encephalitis
Eastern equine encephalitis virus	Alphavirus, Togaviridae	Human, birds, mosquitoes	Zoonosis, arthropod bite	Encephalitis
Ebolavirus	Ebolavirus, Filoviridae	Human, monkeys, bats	Zoonosis, contact	Hemorrhagic fever
Echovirus	Enterovirus, Picornaviridae	Human	Fecal-oral	Common cold
Encephalomyocarditis virus	Cardiovirus, Picornaviridae	Human, mouse, rat, pig	Zoonosis	Encephalitis
Epstein-Barr virus	Lymphocryptovirus, Herpesviridae	Human	Contact, saliva	Mononucleosis
European bat lyssavirus	Lyssavirus, Rhabdovirus	Human, bats	Zoonosis, animal bite	Fatal encephalitis
GB virus C/Hepatitis G virus	Pegivirus, Flaviviridae	Human	Blood, occasionally sexual	None
Hantaan virus	Hantavirus, Bunyaviridae	Human, rodents	Zoonosis, urine, saliva	Renal or respiratory syndrome
Hendra virus	Henipavirus, paramyxoviridae	Human, horse, bats	Zoonosis, animal bite	Encephalitis
Hepatitis A virus	Hepatovirus, picornaviridae	Human	Fecal-oral	Hepatitis
Hepatitis B virus	Orthohepadnavirus, Hepadnaviridae	Human, Chimpanzees	Sexual contact, blood	Hepatitis
Hepatitis C virus	Hepacivirus, Flaviviridae	Human	Sexual, blood	Hepatitis
Hepatitis E virus	Hepevirus, Unassigned	Human, pig, monkeys, some rodents, chicken	Zoonosis, food	Hepatitis

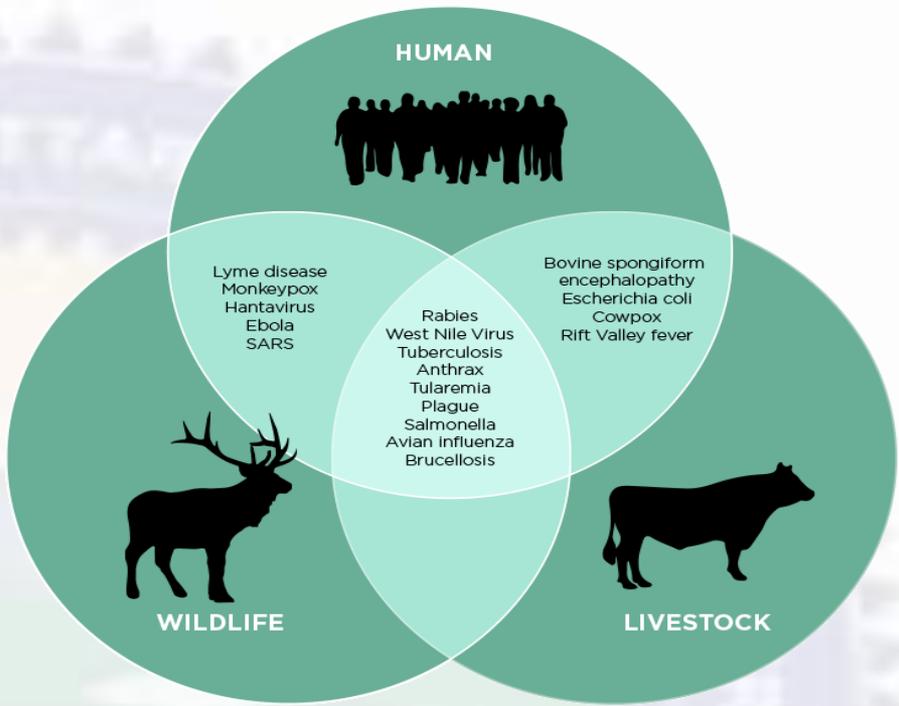
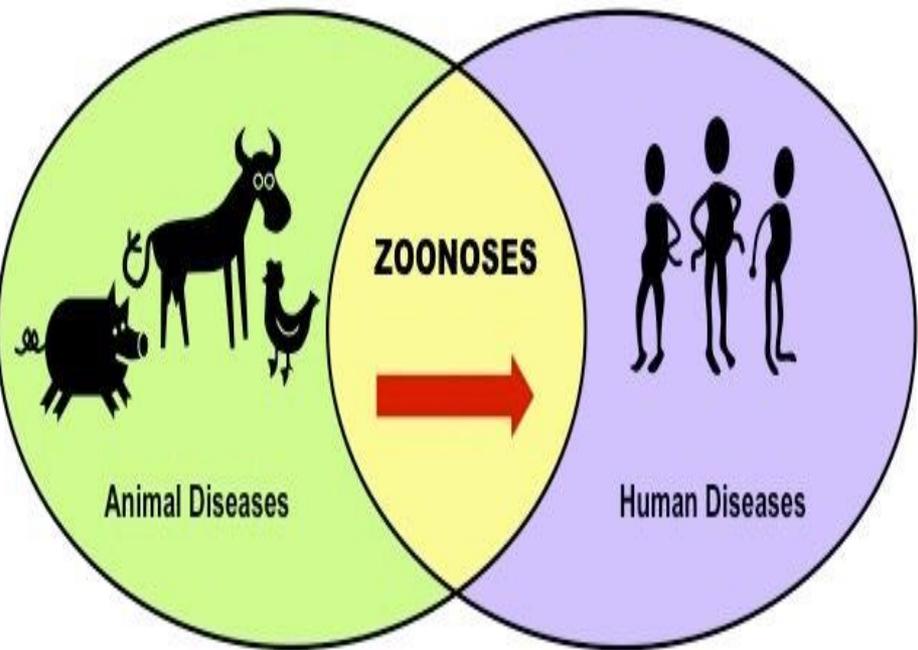
Hepatitis delta virus	Deltavirus, Unassigned	Human	Sexual contact, blood	Hepatitis
Human adenovirus	Mastadenovirus, Adenoviridae	Human	Respiratory, fecal-oral	Respiratory
Human astrovirus	Mamastrovirus, Astroviridae	Human	Fecal-oral	Gastroenteritis
Human coronavirus	Alphacoronavirus, Coronaviridae	Human	Respiratory	Respiratory
Human herpesvirus 1	Simplexvirus, Herpesviridae	Human	Sexual contact, saliva	Skin lesions
Human herpesvirus 2	Simplexvirus, Herpesviridae	Human	Sexual contact, saliva	Skin lesions
Human herpesvirus 6	Roseolovirus, Herpesviridae	Human	Respiratory, contact	Skin lesions
Human herpesvirus 7	Roseolovirus, Herpesviridae	Human	Respiratory, contact	Skin lesions
Human herpesvirus 8	Rhadinovirus, Herpesviridae	Human	Sexual contact, saliva	Skin lymphoma
Human immunodeficiency virus	Lentivirus, Retroviridae	Human	Sexual contact, blood	AIDS
Human papillomavirus 1	Mupapillomavirus, Papillomaviridae	Human	Contact	Skin warts
Human papillomavirus 16,18	Alphapapillomavirus, Papillomaviridae	Human	Sexual	Genital warts, cervical cancer
Human parainfluenza	Respirovirus, Paramyxoviridae	Human	Respiratory	Respiratory
Human parvovirus B19	Erythrovirus, Parvoviridae	Human	Respiratory	Skin lesion
Human respiratory syncytial virus	Orthopneumovirus, Pneumoviridae	Human	Respiratory	Respiratory

Human rhinovirus	Enterovirus, Picornaviridae	Human	Respiratory	Respiratory
Human SARS coronavirus	Betacoronavirus, Coronaviridae	Human, bats, palm civet	Zoonosis	Respiratory
Human T-lymphotropic virus	Deltaretrovirus, Retroviridae	Human	Sexual contact, maternal-neonatal	Leukemia
Human torovirus	Torovirus, Coronaviridae	Human	Fecal-oral	Gastroenteritis
Influenza A virus	Influenzavirus A, Orthomyxoviridae	Human, birds, pigs	Respiratory or Zoonosis, animal contact	Flu
Isfahan virus	Vesiculovirus, Rhabdoviridae	Human, sandflies, gerbils	Zoonosis, arthropod bite	Undocumented, encephalitis?
Japanese encephalitis virus	Flavivirus, Flaviviridae	Human, horses, birds, mosquitoes	Zoonosis, arthropod borne	Encephalitis
Kunjin virus	Flavivirus, Flaviviridae	Human, horses, birds, mosquitoes	Zoonosis, arthropod borne	Encephalitis
Lagos bat virus	Lyssavirus, Rhabdoviridae	Human, mammals	Zoonosis, animal bite	Fatal encephalitis
Lake Victoria marburgvirus	Marburgvirus, Filoviridae	Human, monkeys, bats	Zoonosis, fomite	Hemorrhagic fever
Langat virus	Flavivirus, Flaviviridae	Human, ticks	Zoonosis, arthropod borne	Encephalitis
Lassa virus	Arenavirus, Arenaviridae	Human, rats	Zoonosis, fomites	Hemorrhagic fever
Louping ill virus	Flavivirus, Flaviviridae	Human, mammals, ticks	Zoonosis, arthropod bite	Encephalitis
MERS coronavirus	Betacoronavirus, Coronaviridae	Human, Tomb bat	Zoonosis	Respiratory
Measles virus	Morbilivirus, Paramyxoviridae	Human	Respiratory	Fever, rash

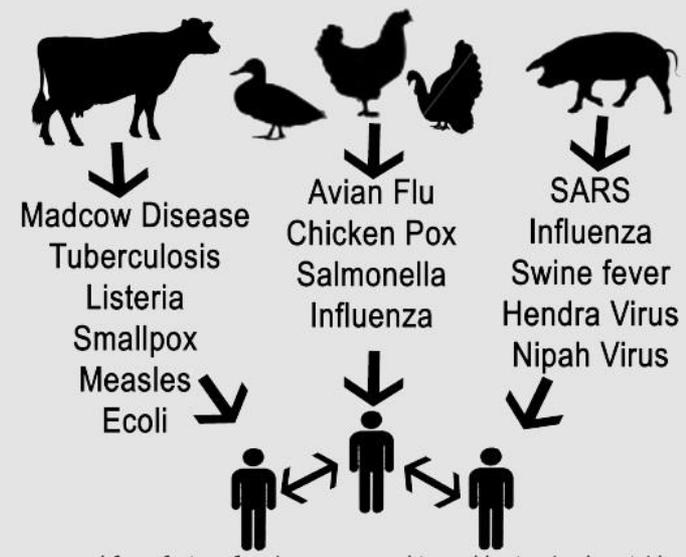
Mengo encephalomyocarditis virus	Cardiovirus, Picornaviridae	Human, mouse, rabbit	Zoonosis	Encephalitis
Mokola virus	Lyssavirus, Rhabdoviridae	Human, rodents, cat, dog shrew	Zoonosis, animal bite	Encephalitis
Monkeypox virus	Orthopoxvirus, Poxviridae	Human, mouse, prairie dog	Zoonosis, contact	Skin lesions
Mumps virus	Rubulavirus, Paramyxoviridae	Human	Respiratory, saliva	Mumps
Murray valley encephalitis virus	Flavivirus, Flaviviridae	Human, mosquitoes	Zoonosis, arthropod bite	Encephalitis
New York virus	Hantavirus, Bunyavirus	Human, mouse	Zoonosis, urine, saliva	Hemorrhagic fever
Nipah virus	Henipavirus, Paramyxoviridae	Human, bats	Zoonosis, animal bite	Encephalitis
Norwalk virus	Norovirus, Caliciviridae	Human	Fecal-oral	Gastroenteritis
O'nyong-nyong virus	Alphavirus, Togaviridae	Human, mosquitoes	Zoonosis, arthropod bite	Fever, joint pain
Orf virus	Parapoxvirus, Poxviridae	Human, mammals	Zoonosis, contact	Skin lesions
Oropouche virus	Orthobunyavirus, Bunyaviridae	Human, wild animals(sloths)	Zoonosis, arthropod bite	Fever, joint pain
Pichinde virus	Arenavirus, Arenaviridae	Human, rat, guinea pig	Zoonosis, fomite	Hemorrhagic fever
Poliovirus	Enterovirus, Picornaviridae	Human, mammals	Fecal-oral	Poliomyelitis
Punta toro phlebovirus	Phlebovirus, Bunyaviridae	Human, sandflies	Zoonosis, arthropod bite	Hemorrhagic fever

Puumala virus	Hantavirus, Bunyavirus	Human, bank vole	Zoonosis, urine, saliva	Hemorrhagic fever
Rabies virus	Lyssavirus, Rhabdoviridae	Human, mammals	Zoonosis, animal bite	Fatal encephalitis
Rift valley fever virus	Phlebovirus, Bunyaviridae	Human, mammals, mosquitoes, sandflies	Zoonosis, arthropod bite	Hemorrhagic fever
Ross river virus	Alphavirus, Togaviridae	Human, mosquitoes, marsupials	Zoonosis, arthropod bite	Fever, joint pain
Rotavirus	Rotavirus, Reoviridae	Human	Fecal-oral	Gastroenteritis
Rubella virus	Rubivirus, Togaviridae	Human	Respiratory	Rubella
Sagiyama virus	Alphavirus, Togaviridae	Human, horse, pig, mosquitoes	Zoonosis, arthropod bite	Fever, joint pain
Sandfly fever sicilian virus	Phlebovirus, Bunyaviridae	Human, sandflies	Zoonosis, arthropod bite	Hemorrhagic fever
Sapporo virus	Sapovirus, Caliciviridae	Human	Fecal-oral	Gastroenteritis
SARS coronavirus 2	Betacoronavirus, Coronaviridae	Human, bats, pangolin?	Respiratory	Covid-19
Semliki forest virus	Alphavirus, Togaviridae	Human, birds, hedgehog, mosquitoes	Zoonosis, arthropod bite	Fever, joint pain
Seoul virus	Hantavirus, Bunyavirus	Human, rats	Zoonosis, urine, saliva	Hemorrhagic fever
Simian foamy virus	Spumavirus, Retroviridae	Human, monkeys	Zoonosis, contact	None
Simian virus 5	Rubulavirus, Paramyxoviridae	Human, dog	Zoonosis, contact	Undocumented

Sindbis virus	Alphavirus, Togaviridae	Human, birds, mosquitoes	Zoonosis, arthropod bite	Pogosta_disease Fe ver, joint pain
Southampton virus	Norovirus, Caliciviridae	Human	Fecal-oral	Gastroenteritis
St. louis encephalitis virus	Flavivirus, Flaviviridae	Human, birds, mosquitoes	Zoonosis, arthropod bite	Encephalitis
Torque teno virus	Alphatorquevirus, Anelloviridae	Human	Sexual, blood	None
Uukuniemi virus	Phlebovirus, Bunyaviridae	Human, ticks	Zoonosis, arthropod bite	Hemorrhagic fever
Varicella-zoster virus	Varicellovirus, Herpesviridae	Human	Respiratory, contact	Varicella
Variola virus	Orthopoxvirus, Poxviridae	Human	Respiratory	Variola
Venezuelan equine encephalitis virus	Alphavirus, Togaviridae	Human, rodents, mosquitoes	Zoonosis, arthropod bite	Fever, joint pain
Western equine Encephalitis virus	Alphavirus, Togaviridae	Human, vertebrates, mosquitoes	Zoonosis, arthropod bite	Fever, joint pain
West Nile virus	Flavivirus, Flaviviridae	Human, birds, ticks, mosquitoes	Zoonosis, arthropod bite	Encephalitis
Yaba monkey tumor virus	Orthopoxvirus, Poxviridae	Human, monkeys	Zoonosis, contact	None
Yellow fever virus	Flavivirus, Flaviviridae	Human, monkeys, mosquitoes	Zoonosis, arthropod bite	Hemorrhagic fever
Zika virus	Flavivirus, Flaviviridae	Human, monkeys, mosquitoes	Zoonosis, arthropod bite	Fever, joint pain, rash

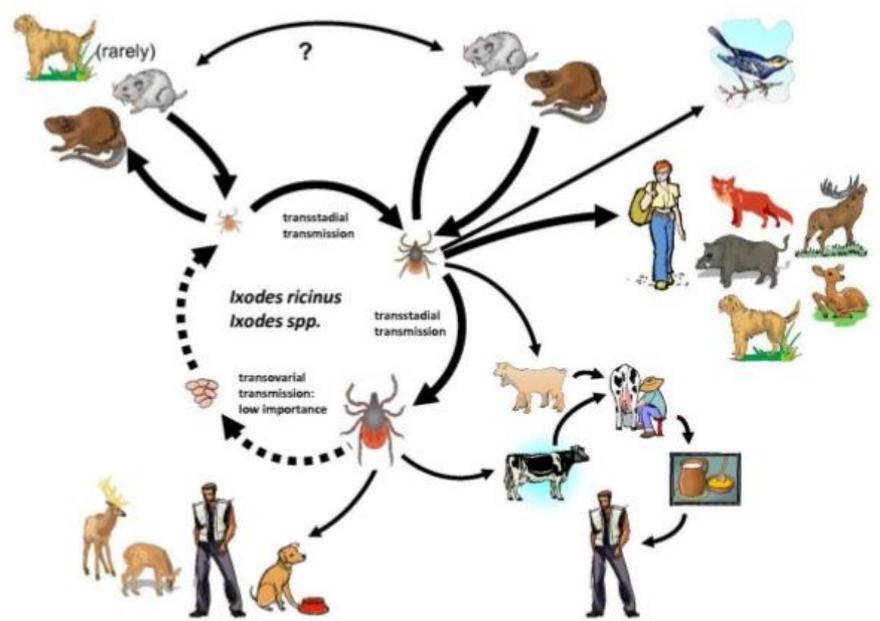


The Animals and the Diseases



-Diseases spawned from factory farming can spread to and hurt animals outside of the farm

TICK BORNE ENCEPHALITIS



Control Measurements of human and animal diseases

1. Prevention of Environmental contamination
2. Control of Intermediate host, vectors and reservoirs
3. Control of internal parasites
4. Control of arthropod pests
5. Control and reducing the infection as soon as an outbreak occurs
6. Isolation of sick animals
7. Quarantine for newly purchased animals
8. Vaccination
9. Deworming of animals
10. Elimination of carriers
11. Tuberculin test for cattle and buffalo
12. Johnin test for cattle and buffalo
13. Agglutination test for brucellosis
14. Test for mastitis-Strip Cup Test
15. Test for mastitis- California Mastitis Test (CMT)
16. Disposal of carcass
17. Burial of carcass
18. Burning of carcass
19. Disinfection of animal houses
20. Disinfection of pastures
21. Common disinfectants and their usage
22. General Disease Prevention Measures

KEY POINTS OF THE LECTURE

- ❑ Viruses are simple and acellular infectious agents.
- ❑ They can infect all types of life forms, from multicellular organisms to unicellular organisms.
- ❑ Viruses are microscopic obligate cellular parasites, generally much smaller than bacteria. They lack the capacity to thrive and reproduce outside of a host body.
- ❑ Regressive Hypothesis, Escaped Gene Theory and Co-evolution Hypothesis explain the origin of viruses.
- ❑ The branch of science which deals with the study of viruses is called virology.
- ❑ The term “virus” is derived from the Latin word *vīrus* referring to poison.
- ❑ In 1892, Ivanovsky first discovered virus in an infected tobacco plant.
- ❑ Martinus Beijerinck called introduced the word virus.
- ❑ The Tobacco Mosaic Virus was the first to be crystallized by Stanley in 1935.
- ❑ Félix d'Herelle described viruses that, when added to bacteria on an agar plate, would produce areas of dead bacteria.
- ❑ Viruses of all shapes and sizes consist of a nucleic acid core, an outer protein coating or capsid, and sometimes an outer envelope.
- ❑ They are devoid of the sophisticated enzymatic and biosynthetic machinery essential for independent activities of cellular life.
- ❑ Viruses are classified into five groups based on morphology.
- ❑ Many viruses attach to their host cells to facilitate penetration of the cell membrane, allowing their replication inside the cell.

KEY POINTS OF THE LECTURE

- ❑ Non-enveloped viruses can be more resistant to changes in temperature, pH, and some disinfectants than are enveloped viruses.
- ❑ The virus core contains the small single- or double-stranded genome that encodes the proteins that the virus cannot get from the host cell.
- ❑ Viral populations do not grow through cell division, because they are acellular.
- ❑ Instead, they use the machinery and metabolism of a host cell to produce multiple copies of themselves, and they assemble in the cell.
- ❑ A virus cannot contain both DNA and RNA. Therefore, virus is called either 'DNA virus' or 'RNA virus'.
- ❑ Viruses are called connective link between living and non living.
- ❑ The main purpose of isolation and purification of viruses is to isolate and identify the virus for clinical specimen and research work.
- ❑ Viruses show lytic or lysogenic mode of life cycle.
- ❑ Transmission of plant viruses may carried through: mechanically, vegetative and graft transmission, pollen transmission, seed transmission, nematode transmission, fungal transmission, insect etc.
- ❑ The definition of a disease is an illness or sickness with specific, well-defined symptoms that affects a person, plant or animal.
- ❑ The diseases may spread through: food, water, air, infection, life style etc.

KEY POINTS OF THE LECTURE

- ❑ Most plant viruses are rod-shaped and rarely have an envelope.
- ❑ The protein discs forming a tube surrounding the viral genome; isometric particles are another common structure in plant virus.
- ❑ Tobacco mosaic virus (TMV), Cucumber mosaic virus (CMV), Barley yellow dwarf virus (BYDV), Tomato spotted wilt virus (TSWV), Prunus necrotic ring spot virus (PNRV), Potato spindle tuber viroid (PSTV), Tomato yellow leaf curl virus (TYLCV) etc are some example of plant viruses.
- ❑ Genetic host resistance, control the vectors, cultural practices, chemical applications and biological control, government regulatory measures etc are some control measurements of the plant viral diseases.
- ❑ Animal viruses may be divided into two class: 1. Vertebrates and 2. Invertebrates.
- ❑ The viruses of vertebrates are causes infections of humans and animals and are called clinical virology and veterinary virology, respectively.
- ❑ Humans cannot be infected by plant or insect viruses, but they are susceptible to infections with viruses from other vertebrates, called viral zoonotic infections.
- ❑ Viruses are important pathogens of livestock and cause diseases such as foot-and-mouth disease and bluetongue.
- ❑ Many viruses, including caliciviruses, herpesviruses, adenoviruses and parvoviruses, circulate in marine mammal populations.
- ❑ Prevention of environmental contamination, control of intermediate host, vectors and reservoirs, isolation of sick animals, quarantine for newly purchased animal, vaccination etc are some important control measurements of the animal viral diseases.

TERMINOLOGY OF VIRUSES

- ❑ **Animal virus:** Any virus capable of infecting one or more animal species.
- ❑ **Assembly:** The gathering and replication of viruses within a cell by using the metabolism of the host organism.
- ❑ **Attachment:** The condition where the capsid proteins of the virus bind to certain receptors of the host organism.
- ❑ **Bacteriophage:** Any virus that infects and replicates within bacteria or archaea.
- ❑ **Capsid:** The outer shell of protein that encloses and protects the genetic material of a virus.
- ❑ **Capsomere:** A subunit of the viral capsid which self-assembles with other capsomeres to form the capsid.
- ❑ **Dalton (Da):** A unit of length frequently used to describe the size of a virus or viral particle.
- ❑ **Envelope:** A lipid casing that surrounds the capsid that covers a virus. A viral envelope assists the virus in infiltrating the cells of the host organism.
- ❑ **Gene Expression:** An activity where information from a gene is made into functional gene material.
- ❑ **Genome Replication:** The reproduction of genetic material, particularly that in the structure of DNA.
- ❑ **Icosahedral:** Having the symmetry of an icosahedron.
- ❑ **Kilobase (kb):** One kilobase is equal to 1000 base pairs.
- ❑ **Latent Infection:** A viral infection that exists in dormancy and does not exhibit symptoms.
- ❑ **Maturation:** The phase during replication at which a virus becomes infectious.
- ❑ **mRNA:** A form of ribonucleic acid which carries copied genetic information from DNA to the cell ribosome.
- ❑ **Neucleocapsid:** The composition of a virus that includes the DNA, RNA, and the capsid protein cover.
- ❑ **Penetration:** The process of the virus entering the cell of the host organism, causing infection.

TERMINOLOGY OF VIRUSES

- ❑ **Receptor:** A specific type of molecule found on a cell membrane that a virus is able to attach to.
- ❑ **Release:** The process of the death of a host cell that discharges a virus.
- ❑ **Replication:** Any of the various processes by which a virus reproduces.
- ❑ **Uncoating:** A condition when the protein capsid of the virus is unsheathed due to enzymes of the cells of the host organism.
- ❑ **Vector:** Insects, such as mosquitoes or ticks, that carry disease from one organism to another.
- ❑ **Viral disease:** Any disease that occurs when an organism's body is invaded by infectious viral particles of one or more pathogenic viruses which attach to, enter, and parasitize susceptible cells.
- ❑ **Viral envelope:** A lipid casing present in some viruses which surrounds the capsid and helps to penetrate the host's cell wall.
- ❑ **Virion:** A singular, stable particle that is the independent form in which a virus exists while not inside an infected cell or in the process of infecting a cell. Virions are the products of a completed viral replication cycle; upon release from the infected cell, they are fully capable of infecting other cells of the same type.
- ❑ **Virions:** A virus particle, which invades the cells of a host organism, causing infection.
- ❑ **Virology:** The study of viruses and virus-like agents, which seeks to understand and explain their structure, classification, evolution, and mechanisms of infection, as well as the diseases they cause, techniques to isolate and culture them, and their use in research and therapy. Virology is often considered a subfield of microbiology or of medical science.
- ❑ **Virus Attachment Protein:** A specific protein found on a virus in charge of fixating to the receptor.
- ❑ **Virus attachment protein:** Any protein which helps to facilitate the binding of a virus to a receptor on a host cell.

SOME QUESTIONS RELATED TO THE LECTURE

- ❑ **Question 1:** What are the viruses? Explain the properties of viruses.
- ❑ **Question 2:** Write a detail note on origin and history of viruses.
- ❑ **Question 3:** Explain the classification of viruses in detail.
- ❑ **Question 4:** Give an illustrated account of the morphology and ultra- structure of the virus.
- ❑ **Question 5:** Write a detail note on the process of isolation and purification of viruses.
- ❑ **Question 6:** What do you understand by replication o viruses? Explain the different stages of life cycle of a virus.
- ❑ **Question 7:** What are the difference between lytic and lysogenic mode of life cycle of a virus.
- ❑ **Question 8:** Define the various mode of transmission of viruses in plants.
- ❑ **Question 9:** Write a detail note on control measurements of viral diseases of plants
- ❑ **Question 10:** Define any plant disease in detail.
- ❑ **Question 11:** What do you understand by term diseases? Describe the various mode of transmission of diseases in vertebrates and invertebrates.

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