LECTURE 2- **lect. Dr. Ahmed Yaseen Abed**

-Nomenclature and Taxonomy of Viruses

-Atypical Virus-like agents (Prions, Defective viruses, Pseudovirion and Viriods)

Classification of Viruses

Classification of viruses is based on the following characteristics:-

1- Virion morphology, including size, shape, type of symmetry, presence or absence of enveloped.

2. Virus genome properties, including type of nucleic acid (DNA or RNA), size of genome, strandedness (single or double), whether linear or circular, positive or negative sense (polarity), segments (number, size).

3.Physicochemical properties of the virion, including PH stability, thermal stability, and susceptibility to physical and chemical agents especially ether and detergents.

4. Virus protein properties, including number, size and functional activities of structural and non-structural proteins, amino acid sequences, and special functional activities (transcriptase,reverse transcriptase, neuraminidase, fusion activities).

5. Genome organization and replication , including gene order, strategy of replication (patterns of transcription, translation), and cellular sites (accumulation of proteins, virion assembly, virion release).

6. Antigenic properties

7. Biological properties, including natural host range, mode of transmission, vector relationships, pathogenicity, tissue tropisms, and pathology.

Nomenclature of viruses

The viruses are classified into groupings which called families, the family names have the suffix-viridae. Each family, subdivided into genera. The genus names carry the suffix-virus.

The name of viruses are derived from:

1. The name of disease caused by virus (eg: influenza virus, hepatitis virus)

2. Organs or tissues they infect with virus (such as; adenovirus)

3. The geographic locality where the virus was **first isolated** (such as; West Nile virus).

4. The name of scientists **responsible for isolating** virus (such as; Epstein- Barr virus)

5. **Unique epidemiological characteristics of virus** (such as: arboviruses; these are arthropod-borne viruses).

Baltimore classification

Viruses were divided into seven groups based on the **their nucleic acid and m-RNA production.**

1- Double strand DNA (**ds-DNA viruses**) for example (adenovirus , herpes viruses) .
2- Single strand DNA **(ss-DNA viruses**) for example (Parvoviruses).
3- **ds- RNA viruses**(e.g. Reo viruses).
4- **(+) ssRNA viruses (+) sense RNA** (e.g. Picornaviruses, Togaviruses).
5- **(-) ssRNA viruses with (-) sense RNA** (e.g. Orthomyxoviruses).
6- **ssRNA-Reverse Transcriptase viruses (+) sense RNA with DNA intermediate** (e.g. Retroviruses)
7- **dsDNA-RT viruses** (e.g. Hepadnaviruses). — Universal system of virus taxonomy: Families – on the basis of virion morphology, genome structure and strategies of replication. Virus family names have the suffix – viridae for example Herpesviridae Genera – based on physicochemical or serological differences.

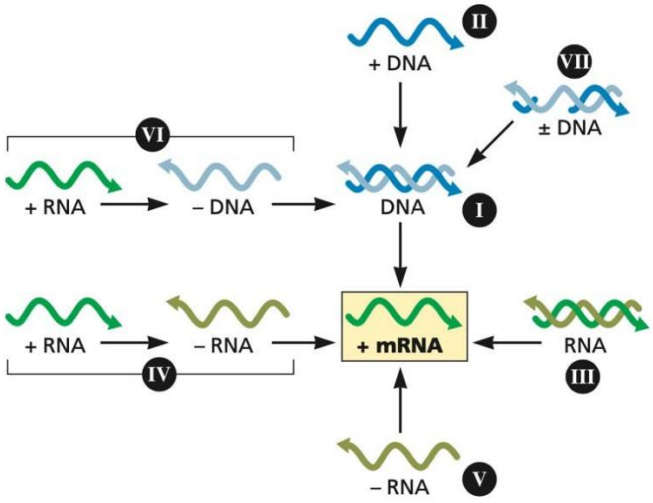


Figure 1.12 The Baltimore classification. The Baltimore classification assigns viruses to seven (I to VII) distinct classes on the basis of the nature and polarity of their genomes. Because all viruses must produce mRNA that can be translated by cellular ribosomes, knowledge of the composition of a viral genome provides insight into the pathways required to produce mRNA, indicated by arrows.

Classification of viruses starts at the level of order and continues as follows with the taxon suffixes given in italics:
Order (*-virales*)
Family (*-viridae*)

Subfamily (*-virinae*)
Genus (*-virus*) Species
Species names generally take the form of [Disease] virus.

Other classification systems are used to classify viruses into different groups.

satellites: small, single-stranded RNA molecules, which lack genes required for their replication. In the presence of a helper virus, they can replicate, there are two types:

a. satellites viruses: most are associated with plant viruses.

b. Satellites nucleic acids (virusoids).

Defective viruses: are composed of **viral nucleic acid** and **protein**, but cannot replicate without co-virus (helper virus) because missing some functions, such as certain adenoviruses and hepatitis D virus) .Defective viruses usually have a **mutation** or a **deletion** of part of their genetic material. During the growth of most human viruses, many more defective than infectious virus particles are produced. The ratio of defective to infectious particles can be as high as **100:1.**

Pseudoviruses: the virus particle contains host cell DNA instead of viral DNA within capsid. They are formed during infection of host cell. Pseudoviruses can infect cells but they don't replicate.

Viroid: consist of only single molecule of circular ssRNA without protein coat or envelope. They replicate and cause several diseases **in plant** but **not** in human. Viroids particles that cause **potato spindle tuber** disease.

Viroids have been found to differ from viruses in six ways:

1. Consists of a single circular RNA molecule of low molecular weight.
2. Exist inside cells, usually inside of **nucleoli** as particles of RNA without capsids or envelopes.

3. Do not require a helper virus.

4. Viroid RNA does not produce proteins.

5. Viroid RNA is always copied in the host cell nucleus.

6. Not apparent in infected tissue without use of special techniques to ID (Identity document) nucleotide sequences in the RNA.

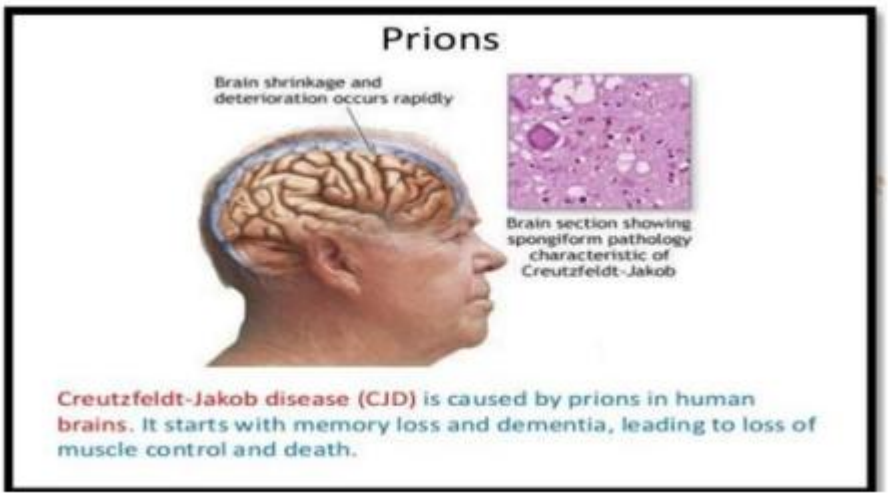
Prion: is infectious particle that is composed only protein. This protein has ability to cause fatal disease. The prion diseases are called **spongiform** (slowly progressive diseases) include: **Scrapie disease** in sheep, **Mad cow** in cattle, and **Kuru disease** in human. Because neither DNA nor RNA has been detected in prions, they are clearly different from viruses. Furthermore, electron microscopy reveals filament rather than virus particles. **Prions** are **much more resistant to inactivation** by **ultraviolet light** and **heat** than are viruses. They are remarkably **resistant to formaldehyde and nucleases**. However, they are inactivated by **hypochlorite**, **NaOH**, and **autoclaving**

Comparison between prions and conventional viruses

Feature	Prions	Conventional viruses
Nucleic acid	No	Yes
Protein	Yes , encoded by cellular genes	Yes ,encoded by viral genes
Heat inactivation	No	Yes
Appearance	Amyloid- like	Icosahedral
Antibody response	No	Yes
Inflammatory responses	No	Yes

Causes of prion disease

Prion diseases occur when **normal prion protein**, found predominantly on the surface of neurons, becomes abnormal and clump in the brain, causing brain damage. This abnormal accumulation of protein in the brain can cause memory impairment, personality changes, and difficulties with movement. Experts still don't know a lot about prion diseases, but unfortunately, these disorders are **generally fatal**.



Q1/ compare between Prion and viroid?

Q2/ Define Pseudovirion , Defective virus?

LECTURE-5
-Immunity & Laboratory Diagnosis of Viruses

A- innate immune-response (non-specific response)

1. **Interferon:** **Alpha** and **beta** interferon are group of proteins produced by human cells after viral infection (have antiviral effects).
   - Inhibit the growth of viruses by **blocking the synthesis of viral proteins**
   - Prevents further spread of viruses
   - Prevents uninfected cells from kill by **NK**

Interferon have two main mechanisms

- **Ribonuclease** that **degrades mRNA**
- **Protein kinase** that **inhibits protein synthesis**

2. **Natural killer cells (NK)** / important part of the **innate immunity** against virus infected cells , called **natural killer cells** because they are active without the necessity of being exposed to the virus previously and they are not specific for any virus

3. **Macrophages** / phagocytosis virus and virus infected cells , production of antiviral molecules such as **INF**

4. **Fever** / elevated body temperature may play a role in host defenses , but it Is important is uncertain

B-Adaptive immune response (Specific response)

1- **Antiviral antibodies** / such as **IgA** confers protection against viruses that enter through **the respiratory and gastrointestinal mucosa**, **IgM** and **IgG** protect against viruses that enter or are **spread through the blood**
2- **Cytotoxic T- lymphocytes** / **CD8+** – positive T cells recognize viral antigen In association with **class I MHC proteins** , they kill virus infected cells by two methods
   - By releasing proteolytic enzymes called **granzymes** into the infected cell , which degrade the cell contents
   - Activating programmed cell death (**apoptosis**)

Diagnosis of Viral Infections

Sampling

A wide variety of samples can be used for virological testing. The type of sample sent to the laboratory often depends **on the type of viral infection** being diagnosed and the test required. Proper sampling technique is essential to avoid potential pre-analytical errors. For example, different types of samples must be collected in appropriate tubes to maintain **the integrity** of the sample and stored at appropriate temperatures (**usually 4°C**) to preserve the virus and prevent bacterial or fungal growth. Sometimes multiple sites may also be sampled.

Types of samples include:

• Blood • Skin • Sputum, gargles and bronchial washings • Urine • Semen

• Faeces • Cerebrospinal fluid • Tissues (biopsies or post-mortem)

• Dried blood spots

Virus isolation

Viruses are often isolated from a patient's first sample. This allows the virus sample to be grown into larger quantities and allows a larger number of tests to be run on them.

This is particularly important for samples that contain new or rare viruses for which diagnostic tests are not yet developed.

Many viruses can be **grown in cell culture in the lab**. Other viruses may require alternative methods for growth such as the **inoculation of embryonated chicken**

eggs (e.g. **avian influenza viruses**) or **the intracranial inoculation of the virus using newborn mice** (e.g. lyssa viruses)

Diagnosis of Viral Infections

Virus diagnosis methods

A-Nucleic acid based methods

Molecular techniques are the most **specific** and **sensitive** diagnostic tests. They are capable of detecting either the whole viral genome or parts of the viral genome. In the past nucleic acid tests have mainly been used as a secondary test to confirm positive serological results. Molecular methods do not rely on the presence of a live virus like virus isolation procedures.

1-Polymerase chain reaction(PCR)

Detection of viral RNA and DNA genomes can be performed using polymerase chain reaction. This technique makes many copies of the virus genome using virus-specific probes. Variations of PCR such as **nested reverse transcriptase PCR** and **real time PCR** can also be used to **determine viral loads** in patient serum. This is often used to monitor treatment success in HIV cases.

2- Sequencing

Sequencing is the only diagnostic method that will provide **the full sequence of a virus genome.** Hence, it provides the most information about very small differences between two viruses that would look the same using other diagnostic tests. For example, sequencing is useful when specific mutations in the patient are tested in order to determine antiviral therapy and susceptibility to infection. However, as the tests are getting **cheaper**, **faster**, and **more automated**, sequencing will likely become the primary diagnostic tool in the future.

B-Microscopy based methods

Immunofluorescence or immunoperoxidase assays are commonly used to detect whether a virus is present in a tissue sample. These tests are based on the principle that if the tissue is infected with a virus, **an antibody specific to that virus will be able to bind to it. To do this,** antibodies that are specific to different types of viruses are mixed with the tissue sample. After the tissue is exposed to a specific wavelength of light or a chemical that allows the antibody to be visualized.

Electron microscopy is a method that can take a picture of a whole virus and can reveal its shape and structure. It is not typically used as a routine diagnostic test as it requires a highly specialized type of sample preparation, microscope and technical expertise.

C-Antibody detection

A person who has recently been infected by a virus will produce antibodies in their bloodstream that specifically recognize that virus. This is **called humoral immunity.** Two types of antibodies are important. The first called **IgM** is highly effective at neutralizing viruses but is only produced by the cells of the immune system for a few weeks. The second, called, **IgG** is produced indefinitely. Therefore, the presence of IgM in the blood of the host is used to test for acute infection, whereas IgG indicates an infection sometime in the past. Both types of antibodies are measured when tests for immunity are carried out.

1-ELISA assay

Antibody testing has become widely available. It can be done for individual viruses (e.g. **using an ELISA assay**) but in **automated panels** that can screen for many viruses at once are becoming increasingly common.

2-Hemagglutination assay

Some viruses attach to molecules present on the surface of red blood cells, for example, influenza virus. A consequence of this is that at certain concentrations a viral suspension may bind together (**agglutinate**) the red blood cells thus preventing them from settling out of suspension.

Questions

1-Mention Laboratory diagnosis of viruses?

2-Enumerate the innate immune-response (non-specific response)? Mention two mechanism of interferon?