

Virus characterization

Aim of virus characterization

- 1- to simplify grouping these viruses and studying their physico-chemical and biological characters.
- 2-for setting viruses indefinite families

Methods of virus characterization

- 1- physical methods
- 2- Chemical methods
- 3- biological methods

Physical characterization

- 1- Electron micro scope
- 2- Exposure to ultraviolet rays
- 3- Heat in activation

1- Electron microscope

- The EM enhanced the discovery of previously unknown microbes that causes previously diseases in man and animal
- Its excellent rapid diagnostic procedures for detection of enteric viruses as Corona& Rota viruses.

2- Exposure to UV rays

- 5ml of the virus suspension is put in sterile petri-dish and put under UV lamp(60 watt)
- The virus infectivity decrease two log or more means that the virus is sensitive to UV rays

3- Heat inactivation

- Some viruses are susceptible to heat at 50c for 30 minutes
- 2ml of the virus suspension in tube and incubate in water bath at 50c for 30minutes
- 2ml of the virus suspension in tube at 4c for 30 minutes
- Then compare the results
- At 50c the virus is inactivated

Chemical methods

- 1- PH stability
- 2- Cationic stabilization
- 3- Essential lipid determination
- 4- Nucleic acid determination

1- PH stability

- Some viruses is inactivated by acidic ph (acid labile)
- While other not affected by acidic ph (acid stable)
- Exposure of viruses to ph3 at 30 minutes leading to decrease their activity as (Human rhino virus)
- While others when exposed to ph3 at 30 minutes not affected as (Human entero virus)

2- Cationic stabilization

- High conc. of divalent cations such as ($MgCl_2$) stabilize certain viruses as (Human enterovirus & Reoviruses) when they are exposed to 50°C for 1 hr)
- While divalent cations increase thermo inactivation of other viruses as (Adenovirus & Herpesviruses)

3- Essential lipid determination

- Lipid solvents remove essential lipids from the nucleocapsid (in naked viruses) and from the envelop (in enveloped viruses) so decreasing of the virus infectivity

Essential lipid determination

- 1- Chloroform sensitivity
- 2- Ether sensitivity
- 3- Deoxy cholate

1- Chloroform sensitivity

- Mix chloroform with virus suspension at 4c for 10m
- Centrifugation at 5000rpm/5m
- Remove the upper aqueous layer and titrate for virus infectivity
- Another sample with out adding chloroform
- The result is decreasing the viral infectivity one log or more in treated sample indicate the susceptibility to chloroform

2- Ether sensitivity

- 4 parts of the virus+ 1 part of ether at 4c for 24hrs
- Remove the upper aqueous layer and titrate for virus infectivity
- Another sample with out adding ether
- The result is decreasing the viral infectivity one log or more in treated sample indicate the susceptibility to ether

3- Deoxy cholate sensitivity

- **Materials:**
- Sodium deoxy cholate solution with ,75 % bovine albumin
- Cell culture for viral titration
- Phosphate buffer at ph7

- **Procedure:**
- Prepare ,2% sodium deoxy cholate solution with ,75% bovine albumin
- This solution stored at 4c but should be warmed to 37c before using
- Centrifugation viral suspension at 5000rpm/1hr
- Mix 1 part of clarified virus suspension with 1 part of ,2% sodium deoxy cholate sodium

- Using viral suspension without sodium deoxycholate as control
- Incubate the mixture at 37c for 1hr
- Adding PBS to the mixture at 4c for 24hr to remove deoxycholate as its toxic to the cell culture
- Titration for viral infectivity
- The result is decreasing the viral infectivity one log or more in treated sample indicate the susceptibility to deoxycholate

4- Nucleic acid determination

- Occur by extraction of nucleic acid
- Procedure of NA extraction:
 - 1- sample preparation
 - 2- protein digestion
 - 3- NA extraction
 - 4- NA preparation
 - 5- detection of NA purity and concentration

Thank you