

VIRAL LABORATORY INSTRUMENTS AND SAFETY CABINETS

Khder Hussein Rasul Zahra Mohammad Maghded Practical Medical Virology and MA 403 Summer Term First week 12/08/2024



Outline

- Viral laboratory instruments
- Biosafety levels
- Biosafety cabinets



Objectives

- To learn about viral laboratory instruments
- $\circ~$ To learn about biosafety levels and differentiate between them
- To learn about biosafety cabinets and differentiate between them

Instruments

Instruments which are used in virology lab are the followings

- 1. Autoclave
- 2. Refrigerator or laboratory freezer
- 3. Microscope
- 4. Centrifuge
- 5. Cytocentrifuge
- 6. Oven
- 7. Filtering unit
- 8. Cryostat
- 9. Incubators
- **10. PCR machine**
- 11. Plate reader
- 12. Electrophoresis equipment
- 13. The single and multichannel micropipettes
- 14. Biosafety cabinets



Autoclave



Autoclave is a device depends on a steam sterilization; viruses, bacteria and fungal spores are killed at 121 °C within 15-20 min and

1.5 bar of pressure of autoclave.



Refrigerator or laboratory freezer, microscope and centrifuge



Refrigerator: Laboratory freezer

The primary function of a laboratory refrigerator is to maintain a defined, internal storage temperature (typically in the +1°C to +15°C range or -10 to -80) in order to provide secure storage and protection of temperature-sensitive products, samples, specimens, chemicals, drugs, solutions, and other substances.

Microscopes are essential for observing viruses and infected cells.

 \circ Electron microscopes (TEM and SEM) are often used for high-resolution imaging of viruses.

Centrifuge is a device that uses centrifugal force to separate various components of a fluid.

- Normal centrifuge (ordinary centrifuge)
- Ultra-centrifuge: it is used at high speeds 45000-60000 rpm
- Micro centrifuge: used for spinning liquid samples as low as 2 ml or even less at high speeds

Cytocentrifuge



Cytocentrifuge: sometimes referred to as a **cytospin**, is a specialized centrifuge used to concentrate cells in fluid specimens onto a microscope slide so that they can be stained and examined.

• The method can be used on many different types of specimens, including fine needle aspirates, cerebrospinal fluid, serous and synovial fluid, and urine.





Oven



Dry heat (oven): It is done in a special instrument (dry oven), which is heated electrically to a temperature of 160 °C for 1-2 hrs.



 \circ Ovens are used to sterilize items.

• Sterilization is defined as the killing or removal of all microorganisms, including bacterial spores.

Filtering unit



In virology, **filtration** refers to a laboratory technique and process used to separate and purify viral particles from other components in a sample, such as cell debris, proteins, nucleic acids, and other contaminants.

• Solid particles that cannot pass through the filter medium are described as **oversize** and the fluid that passes through is called the **filtrate**.



Refrigerated microtome (Cryostat)



Cryostat is a microtome machine for cutting tissue at low temperatures (typically around -15 to -30° C). it is used to make sections. The frozen piece of tissue is placed on the chuck of the refrigerated microtome and cut into sections of the desired thickness





Incubator, PCR machine, plate reader and electrophoresis equipment

Incubators: Incubators maintain controlled temperature and humidity conditions for the growth and propagation of viruses in cell cultures.

PCR Machines: Polymerase Chain Reaction (PCR) machines are used for amplifying viral genetic material, which is essential for virus detection, quantification, and characterization.

Plate readers are used to perform various assays, such as enzyme-linked immunosorbent assays (ELISAs) for virus detection and quantification.

Electrophoresis equipment: Gel electrophoresis is used to separate and analyze viral nucleic acids and proteins based on their size and charge.

The single and multichannel micropipettes



Micropipettes is a precision laboratory instrument used for accurately measuring and transferring small volumes of liquid.



Instructions for pipetting liquids using a micropipette

Micropipettes have 3 positions:

- a) Rest position
- b) First stop
- c) Second stop

The single and multichannel micropipettes





Instructions for pipetting liquids using a micropipette

Micropipettes have 3 positions:

- a) Rest position
- b) First stop
- c) Second stop

Safety manual

Every laboratory should have a safety manual.

A list of some of the safety issues to be addressed in a laboratory safety manual

- 1. Fire safety
- 2. First aid
- 3. Safe use of laboratory glassware
- 4. Safety with handling boiling liquids
- 5. Cleaning up spills and broken glass
- 6. Decontamination of benches and waste
- 7. Recycling of infected materials for example, needles and syringes
- 8. Safe use of chemicals

Biosafety levels



Biosafety levels (BSL) are a set of laboratory containment and safety measures designed to protect laboratory personnel, the environment, and the community when working with potentially hazardous biological materials.

• Once risk is assessed then the appropriate BSL is determined.

• The levels of containment range from the lowest biosafety level 1 (BSL-1) to the highest at level 4 (BSL-4), which includes:

- 1. BSL-1
- 2. BSL-2
- 3. BSL-3
- 4. BSL-4

Biosafety levels



BSL-1 laboratories are used for basic research and teaching activities involving well-characterized and low-risk agents that are not known to cause disease in healthy humans.

BSL-2 laboratories are used for handling agents that pose a moderate risk to personnel and the environment. These agents may cause diseases in humans but have available treatments or vaccines.

BSL-3 laboratories are used for work with agents that can cause severe or potentially lethal diseases in humans, such as tuberculosis, anthrax, and severe acute respiratory syndrome coronavirus (SARS-CoV).

BSL-4 laboratories are the highest level of containment and are used for work with the most dangerous agents, including Ebola virus, Marburg virus, and other highly contagious and deadly pathogens.

Biosafety cabinets



A biosafety cabinet (BSC), also known as a biological safety cabinet or microbiological safety cabinet, is a specialized piece of laboratory equipment designed to provide a controlled and safe working environment for handling hazardous biological materials, including bacteria, viruses, and fungi.

• Biosafety cabinets are essential in research, clinical, and diagnostic laboratories to protect

- 1. Laboratory workers
- 2. The environment
- 3. The integrity of the experiments



- 1. Laminar flow clean air cabinet Class I: This protects the work against microbial contamination by a laminar flow of sterile air.
- This type of cabinet is suitable for making media and handling of non-infected tissue cultures. It is not suitable for infectious work as personnel are exposed to the exhaust air.







2. Class II microbiological safety cabinet: This protects the worker. Air is drawn from the room, with the air steam away from the worker thus protecting him against infection. The incoming air is not sterile and may contaminate the work. The exhausted air is filtered to protect the environment.



- 1- Open front with inward airflow
- 2- Working space
- 3- Exhaust fan
- 4- Exhaust hepafilter
- 5- Sterile exhaust air







3. Class III microbiological safety cabinet: This protects both the work and the worker. Incoming and exhaust air is filtered. The open front is protected by an air current, which acts as a curtain.



Open front with air curtain
Window
Working space
Inlet with hepafilter
Inlet fan
Air inlet with prefilter
Air vents in the bottom
Exhaust fan
Exhaust hepafilter







4. Class IV bacteriological safety cabinet: This type gives maximal protection for the test, the worker, and the environment. Regular inspection of the filter and the fan motors is necessary for maintaining the maximal safety level.



9 and 10- A double set of hepafilters in the exhaust



References (in APA style)

- Arie J. Zuckerman, Jangu E. Banatvala, Paul Griffiths, Barry Schoub, Philip Mortimer Principles and Practice of Clinical Virology-Wiley (2009).
- John Carter, Venetia Saunders Virology Principles and Applications-Wiley (2007).
- A. Zuckerman, et al. Principles and Practice of Clinical Virology-Wiley (2004).