COVENANT UNIVERSITY
NIGERIA

TUTORIAL KIT
OMEGA SEMESTER

PROGRAMME: MICROBIOLOGY

COURSE: MCB 222

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MCB 222: Techniques in Virology

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1. Visualization of virus particle requires the use of electron microscope and is impossible with a light microscope, why?

   Virus particles are very minute in size requiring to be very highly magnified to be seen. The shorter wavelength of electrons (shorter than the wavelength of light used in the light microscopes) increases the resolution ability of the electron microscope thus empowering the electron microscope to produce clear images even at the super high magnification required to view minute viruses. State five virus diseases that constitute serious health concerns around the world.

2. State the essential components of an electron microscope and state the major function of each component

   The essential components of all electron microscopes include the following:
   - **Electron Source ("Gun") produces a stream of electrons**
   - **Electron Lenses (condenser, objective, intermediate and projector lenses). Condenser lenses 1 and 2 focuses the stream of electrons into a small, thin, coherent beam whiles the intermediate and projector lenses, enlarge the image.**
   - **Sample Stage. After mounting on a grid or stub, the specimen is then placed on the stage for viewing.**
   - **Detectors for all signals of interest. For detecting useful signals for image formation.**
   - **Display/Data output devices. For viewing the image.**

3. In your view, what is the most important step in virus replication and why?

4. State one important feature of each of the three types of cell lines and indicate the frequency of passaging each can survive.

   a. Primary cell culture
      - **Normal cells obtained from fresh organs of animals.**
      - **Cannot be maintained in serial culture**
      - **Commonly used for primary isolation of viruses and in preparation of vaccine**
      - **They can be passaged only once or twice**

   b. Diploid or semi continuous cell lines
      - **Subsequent cultures derived from primary cell cultures.**
      - **Cells of single type with diploid chromosome number**
      - **Rapid growth rate**

- **Used for isolation of some fastidious viruses and production of virus vaccines**
- **Can be passaged 50 serial times**

c. Continuous cell lines

- **Derived from a single separated cell thus are called as clones (Cells of a single type)**
- **Usually derived from the cancer cells**
- **Grows fasts**
- **Chromosomes are haploid.**
- **Capable of continuous passaging indefinitely**

5. A diagnostic virology laboratory must be designed for Biosafety level ……. or above. State the characteristics of microorganisms that require the stated biosafety level.

6. State five important things that you would put in place to ensure adequate functionality of your organization's virology laboratory.

7. State two differences between the light microscope and the electron microscopes.

8. State two differences between the transmission electron microscope and scanning electron microscopes.

TEM	SEM
• **Electron beam passes through thin sample.**	• **Electron beam scans over surface of sample.**
• **Specially prepared thin samples or particulate material are supported on TEM grids.**	• **Sample can be any thickness and is mounted on an aluminium stub.**
• **Specimen stage halfway down column.**	• **Specimen stage in the chamber at the bottom of the column.**
• **Image shown on fluorescent screen.**	• **Image shown on TV monitor.**
• **Image is a two dimensional projection of the sample.**	• **Image is of the surface of the sample.**
• **Requires training and some level of skill to operate.**	• **Comparatively easy to operate, with user-friendly interfaces.**
• **Require specialized sample preparation.**	• **Require minimal sample preparation.**

9. State five pieces of equipment and/or materials that you would require for the following procedure and in each case, state the function of the stated equipment or material;
    a. Virus cultivation in laboratory animal.
    b. Virus cultivation in embryoneted egg.
    c. Virus cultivation in cell/tissue culture.
    d. Virus cultivation in whole plant.
    e. Sample preparation for transmission electron microscope.

f. Sample preparation for scanning electron microscope.

10. Indicate the components of a virus targeted in (i) Immunoassays (ii) Molecular tests. Give two examples of each test type.
    (i) Virus coat protein e.g. enzyme linked immunosorbent assay (ELISA), Western blotting, dip stick tests
    (ii) Virus nucleic acid e.g. Polymerase chain reaction (PCR), Southern blotting, Northern blotting.

11. What is serology?
Answer: This is the use of serum antibodies to detect and measure antigens, or the use of antigens to detect serum antibodies.

12. What is the principle of serological diagnosis?

    Answer: Reactions of antigens and antibodies are highly specific as an antigen will react only with antibodies elicited by itself or a closely related antigen. During most infectious diseases, antibodies and immune cells specific for the infecting agents are produced. Detection and measurement of such antibodies aid in the diagnosis and management of such diseases.

13. What are the limitations of serological diagnosis?

14. The complement fixation test is based on what principle?

15. What are the essential components of the complement fixation test?
    Answer: Patients serum containing specific antibodies; sheep red blood cells; hemolysin; complement and antigens.

16. What is immunofluorescence?
    Answer: Immunofluorescence is the labelling of antibodies or antigens with a fluorescent dye in order to identify or locate them in a tissue sample.

17. Differentiate between direct and indirect immunofluorescence.

18. Mention two instances in which neutralization tests are used in serological diagnosis.

19. What is haemagglutination?
    Answer: Haemagglutination is the clumping of red blood cells.

20. What specimens are tested in serological diagnosis?