### Shivaji University, Kolhapur Yashwantrao Chavan College of Science Karad Department of Microbiology

Class - M.Sc. I, Subject Name: Techniques in Microbiology

#### True / False type questions

- 1. Dark field microscope produces a bright image of the object against a dark background.
- 2. Dark field microscope enhances the phase contrast between intracellular structures having slight differences in refractive index.
- 3. Differential Interference contrast microscope (DIC) creates images by detecting differences in refractive indices & thickness of different parts of the specimen.
- 4. In a DIC microscope specimens are stained with fluorochrome.
- 5. Fluorescent microscope shows a bright image of the object resulting from the fluorescent light emitted from a specimen.
- 6. Objects having size between 100μm to 1A<sup>0</sup> can be observed by using an electron microscope.
- 7. In an electron microscopy electrons are emitted by a heated filament of Tungsten.
- 8. In an electron microscopy electrons are emitted by a cold filament of tungsten which acts as an anode.
- 9. Fluorescent microscope shows a bright image of the object resulting from the electrons emitted from a specimen.
- 10. Laser beam is used to illuminate the spots on a specimen in case of confocal microscopy.
- 11. Hungate roll tube technique employed in an attempt to cultivate a minimal portion of the organism in the gingival crevice area of man.
- 12. Sodium thioglycolate is an example of an oxidizing agent in the cultivation of anaerobes.
- 13. Sodium thioglycolate is an example of a reducing agent in the cultivation of anaerobes.
- 14. Many specimens are too thick to be mounted directly onto a slide, and hence these are cut into thin sections using a device called acrotome.
- 15. Rhodamine is an example of flurochrome used in FISH.
- 16. Enrichment culture technique is the use of certain growth media to favour the growth of a particular bacterium over the others.
- 17. Laboratories with short-time cultivations follow the techniques like sub-cultivation, storage under mineral oil and by cooling to  $4-8^{\circ}$ C.
- 18. Sporulating fungi stored for 7-18 years in silica gel and can remain morphologically stable.
- 19. Silica gel storage is a technique that involves the inoculation of a suspension of fungal propagules onto a cold silica gel.
- 20. In a lyophilization technique recrystallization of ice can occur at temperatures above -139°C and this can cause a structural damage during storage.
- 21. Good Laboratory Practices was first introduced in New Zealand and Denmark in 2009.
- 22. Good Laboratory Practices is a formal regulation created by USFDA.
- 23. Good Laboratory Practices were initially implemented to prohibit hygenic environment in the laboratories.
- 24. Animal care facilities should be located away from the testing laboratories.
- 25. If a personnel is exposed to acids and alkalies, such type of hazard is called biological hazard.
- 26. Contamination risk is reduced by barrier system.



- 27. Reagents used in the operation should be specified in the SOPs.
- 28. Archieves should be there for orderly storage and expedient of all documents.
- 29. GLP is a FDA regulation which is accepted and approved as international standards by OECD.
- 30. Exposure of personnel to contaminated body fluids is a biological type of hazard.
- 31. Risk assessment of each laboratory task is conducted by OSHA.
- 32. The full form of ACDP is Advisory Committee on Dangerous pathogens.
- 33. The first thing for implementation of action plan for safety practices is to access level
- 34. In case, if personnel exposed to a hazard, one should immediately report the incidence.
- 35. In a laboratory, alcohol used for disinfection of hands is of 34% concentration.
- 36. Wrappers should be dumped of in different dust bins and not mixed with normal domestic waste.
- 37. PPO kit should be used while handling highly transmissible pathogen.
- 38. For general laboratory disinfection, hypochlorite solution should be diluted at 1% concentration.
- 39. Ultrasonication method is used for cell disruption.
- 40. Cation exchange chromatography retains cations and has negatively charged stationary phase functional group.
- 41. Conventional analytical columns in GC usually use flow rates in the range from 20-50 mL/min.
- 42. In SDS PAGE, one SDS molecule binds for every four amino acid residues.
- 43. Gels containing 0.3% agarose will separate double-stranded DNA molecules of between 5 and 60 kb size.
- 44. The polymerization of acrylamide is an example of free-radical catalysis.
- 45. If one is aiming to detect a particular protein often an enzyme on the basis of its biological activity, SDS PAGE is used.
- 46. For capillary electrophoresis, reagents are required at larger quantity.
- 47. Sample for agarose gel electrophoresis are in a buffer solution that contains ficoll.
- 48. Differential centrifugation is based upon the differences in the sedimentation rate.
- 49. Percoll is gradient material used in density gradient centrifugation.
- 50. The target analyte retained on the stationary phase can be eluted by increasing the concentration of a similarly charged species.
- 51. The most frequently used capillary column in GC, is the fused silica open tubular
- 52. Photo-polymerisation is an alternative method that can be used to polymerise acrylamide gels.
- 53. High percentage of epichlorohydrin gives large pore size for sephadex.
- 54. Stationary phase in GLC is a solid like silica or alumina.
- 55. The carrier gas pressure ranges from 1-5 psi.
- 56. The most frequently used capillary column is the fused support coated open tubular.
- 57. In cation exchange chromatography, the negatively charged analyte could be displaced by the addition of positively charged sodium ions.
- 58. Columns used in HPLC should withstand high pressure of up to 1.5 x 10<sup>7</sup> Pa.
- 59. In isocratic elution, separation of target analyte is done by using more than two solvent.
- 60. In photo-polymerisation ammonium persulphate are replaced by riboflavin.
- 61. SDS is cationic detergent.
- 62. Stacking gel is poured on top of the separating gel contain 14% acrylamide.
- 63. Pure protein gives two bands on an SDS-PAGE, unless the molecule is made up of two unequal subunits.

- 64. In native gels, polyacrylamide gels are used normally at concentration of 7.5%.
- 65. Much greater range of protein with Mr values can be separated on a fixed-percentage gel.
- 66. Bio-Lyte is an example of ampholyte used in isoelectric focusing.
- 67. Fluorescence is an emission phenomenon where an energy transition from a lower to a higher state is accompanied by radiation.
- 68. Proteins possess three intrinsic fluorophores as tryptophan, tyrosine and phenylalanine.
- 69. Bioluminescence is the reaction leading to a fluorescent product which involves enzyme in luciferase.
- 70. The main application of bacterial luciferase is the determination of electron transfer co-
- 71. Luminometry essentially measures the angle through which the plane of polarisation is changed.
- 72. Optically active molecule has a positive CD then its enantiomer will have a negative CD
- 73. The absorption of infrared light by a molecule results in transition to higher levels of vibration.
- 74. The criterion for a peak to appear in the Raman spectrum is a change in polarisability of the molecule.
- 75. NMR spectroscopy is the main method of structure determination for organic compounds.
- 76. The basic principles of NMR can be applied to imaging of dead samples.
- 77. Some isotopes decay by emitting positively charged b-particles referred to as positrons.
- 78. An alpha particle is a helium nucleus which consists of one proton and one neutron.
- 79. Radioactive decay is measured as destructions per minute.
- 80. The International Unit of the radioactivity is Becquerel.

## Long answer type questions. Each question carries 16 marks.

- 1. Discuss in detail general principles of isolation and cultivation of anaerobes.
- 2. Describe in detail principles and working of transmission and scanning microscopes.
- 3. Explain in detail general principles, methods and selective factors used in enrichment culture technique.
- 4. Discuss in detail principles and methods of preservation of bacteria, viruses, yeasts and molds.
- 5. Describe in detail principles and working of dark field and phase contrast microscopy.
- 6. Explain in detail principles and methods of cell disruption.
- 7. Explain in detail general principles of Good laboratory practices.
- 8. Explain in detail common hazards in the laboratory.
- 9. Discuss in detail safety measures used in the microbiology laboratory.
- 10. Explain in detail general principle, working and applications of ion exchange chromatography.
- 11. Explain in detail general principle, working and applications of gas chromatography.
- 12. Explain in detail general principle, working and applications of high performance liquid chromatography.
- 13. Give the detailed account of principle, working and applications of polyacrylamide gel electrophoresis.
- 14. Explain in detail SDS polyacrylamide gel electrophoresis. Add note on its applications.
- 15. Give the detailed account of principle, working and applications of agarose gel electrophoresis.

MICHOBIOLOG

- 16. Describe in detail general principles of radioisotopic techniques. Add a note on GM counter.
- 17. Explain in detail principles of IR and Raman spectrophotometry.
- 18. Describe in detail different methods of using radioisotopes.
- 19. Describe in detail different methods of detection of radioactivity.
- 20. Give the detailed account of principle, working and applications of X ray crystallography.

### Medium answer type questions (Each question carries 08 marks)

- 1. Give principles and methods of preservation of viruses.
- 2. Give principles and methods of preservation of yeast and molds.
- 3. Describe in detail principles and working of transmission microscopes.
- 4. Describe in detail principles and working of scanning microscopes.
- 5. Explain in brief reducing agents and indicators used in cultivation of anaerobic bacteria.
- 6. Give principles and methods of preservation of bacteria.
- 7. Explain in detail principles and selective factors used in enrichment culture technique.
- 8. Methods and media used for the isolation of human and animal pathogenic fungi.
- 9. Describe in brief principles, working and applications of dark field microscopy.
- 10. What is Good laboratory practice? Explain GLP in microbiology laboratory.
- 11. What is safety? Discuss various safety measures used in the laboratory.
- 12. Discuss in brief qualifications of equipment.
- 13. Explain in brief validation and calibration.
- 14. Discuss in brief methods of disruption of microbial cells.
- 15. Discuss in brief methods of disruption of plant and animal cells.
- 16. Explain in brief common chemical hazards in the laboratory.
- 17. Explain in detail working and applications of gel exclusion chromatography.
- 18. Explain in brief SDS polyacrylamide gel electrophoresis.
- 19. Discuss in detail principle and working of differential and density gradient centrifugation.
- 20. Explain in detail types of gas chromatography and add note on components of gas chromatography.
- 21. Explain in detail working and applications of capillary gel electrophoresis.
- 22. Discuss in brief working mechanism of ion exchange chromatography.
- 23. Explain in brief nature, types and characteristics of gels used in gel electrophoresis.
- 24. Explain in brief general principles of column chromatography.
- 25. Give general principles of electrochemical cells and potentiometry
- Explain in brief principles and applications of circular dichroism and optical rotational dichroism.
- 27. Describe in detail any two methods of detection of radioactivity.
- 28. Explain in detail principle and working of Raman spectrophotometry.
- 29. Explain in detail principles and applications of IR spectrophotometry,
- 30. Explain in brief principles and applications of the pH and oxygen electrodes.

# Short answer type questions (Each question carries 04 marks).

- 1. Hungate's roll tube technique
- 2. Open enrichment system.
- 3. Differential interference contrast
- 4. Single cell isolation
- 5. Preservation of yeast and mold

- 6. Closed enrichment system
- 7. Preservation of bacteria
- 8. Preservation of viruses
- 9. Reducing agents and indicators used in anaerobic media
- 10. Anaerobic jar method
- 11. Anaerobic glove box
- 12. Media used for isolation of human pathogenic fungi
- 13. Atomic force microscopy
- 14. Confocal scanning
- 15. Selective factors used in enrichment technique
- 16. Performance qualifications
- 17. Design qualifications
- 18. Installation and operational qualifications.
- 19. Types and significance of validation.
- 20. Accuracy in preparation of solutions and media.

- 21. Ionizing radiations as hazards
- 22. Infectious materials as hazards
- 23. Gas and fire hazards
- 24. Personal protection in laboratory
- 25. Waste disposal
- 26. Use of First aid box in laboratory.
- 27. Concept of Documentation
- 28. Need and types of Documentation.
- 29. Microbial cell disruption.
- 30. Plant cell disruption.
- 31. Applications of gel exclusion chromatography.
- 32. Isoelectric focusing
- 33. Density gradient centrifugation
- 34. Native and gradient gels
- 35. Electrophoresis of RNA
- 36. Diagrammatic representation of capillary electrophoresis
- 37. Applications of ion exchange chromatography
- 38. Cation and anion exchangers
- 39. Detectors used in HPLC
- 40. Carrier gas used in gas chromatography
- 41. Gas chromatography columns
- 42. Formation of Polyacrylamide gels.
- 43. Applications of SDS-PAGE
- 44. Two-dimensional polyacrylamide gel electrophoresis
- 45. Pulsed-field gel electrophoresis
- 46. Turbidimetry
- 47. Nephelometry
- 48. Fluorimetry
- 49. Luminometry
- 50. Circular dichroism
- 51. Optical rotational dichroism
- 52. Spectrophotometry
- 53. ESR
- 54. NMR
- 55. Gas ionization chamber



- 56. Radioisotope tracer technique,
- 57. Isotope dilution assay
- 58. pH electrode
- 59. Ion selective electrode
- 60. Oxygen electrodes