(1)

Q.P. Code: 67147

S.Y.B.Sc. Biotechnology Semester IV (Revised) Examination

Biotechnology- Molecular diagnostics

Model Answers

Q 1	**Do as directed (Any fifteen)**	**15**
1.	True	
2.	True	
3.	In Southern blotting, the doublestranded DNA fragments in the gel must be denatured and transferred to a **nitrocellulose** membrane	
4.	**Restriction endonucleases are also called as restriction enzymes which recognizes a specific nucleotide-pair sequence in DNA called a restriction site and cleaves the DNA (hydrolyzes the phosphodiester backbones) within or near that sequence.**	
5.	Fill in the blank: Proteins are separated using **SDS-polyacrylamide gels (SDS-PAGE) or isoelectric focusing gels (IEF)**	
6.	Fill in the blank: Taq DNA polymerase enzyme from ***Thermus aquaticus*** has facilitated the process of molecular diagnostics.	
7.	In paternity testing, if a man lacked the allele that must have come from the baby's father, then the DNA typing data would have proved that he is not the father; this is known as **Exclusion** result.	
8.	Give an example of chelator present in master mix. **EDTA**	
9.	The type of PCR control which ensures that the enzyme is active, the buffer is optimal, the primers are priming the write sequences, and the thermal cycler is cycling appropriately is called as _____. **Positive control**	
10.	_____ is used to increase the effectiveness of UV light to decontaminate and maintain pre-PCR area. **Psoralens**	
11.	The *Tth* polymerase is isolated from _____. ***Thermus thermophilus***	
12.	Give an example of: Monovalent cations present in PCR master mixture **KCl**	
13.	Define the term 'amplicons'. **An amplicon is a piece of DNA or RNA that is the source and/or product of amplification.**	
14.	What is the significance of thermocyclers? **Thermocyclers are designed to rapidly and automatically ramp (change) through the required incubation temperatures, holding at each one for designated periods.**	
15.	Presence of quasi species of HIV is due to the error-prone nature of the reverse transcriptase enzyme. State true or false **True**	
16.	It is the time period between exposure to HIV and before seroconversion is known as _____. **Window period**	

17.	Pace-2 is a second generation hybridisation assays used for *N.gonorrhoeae*. State true or false **False**	
18.	_____gene analysis is done to detect fragile X diagnostic. **FMR1 gene**	
19.	Intrapartum transmission of *N.gonorrhoeae* produces _____ condition in neonates. **Conjunctivitis or Ophthalmia neonatorum**	
20.	_____ is the process of determining an individual's DNA characteristics, which are as unique as fingerprints. **DNA profiling/ DNA fingerprinting**	
Q. 2 A	What is molecular diagnostics? Explain in detail the of mutation detection techniques in molecular diagnostics Definition (1Mark) Significance of PCR (2 Marks) Techniques developed – Enzymatic based methods, Electrophorectic based method, Solid phase based techniques (5Marks)	**08**
Q. 2 B	Elaborate on FISH as a hybridisation technique. FISH- Full form 1 mark Technique and Applications – 6 marks	**07**
	OR	
Q. 2 C	Discuss the organic and inorganic method for isolation of DNA. Organic DNA isolation method - Diagram (1 Mark) Steps and significance of phenol chloroform mixture (3 Marks) Inorganic DNA isolation method – Diagram (1 Mark) Salting out method (3Marks)	**08**
Q. 2 D	Explain western blotting in detail. The immobilized target for a Western blot is protein (1Mark) Separation using SDS-polyacrylamide gels (SDS-PAGE) or isoelectric focusing gels (IEF) (1Mark) Significance of chemicals used and blocking(3Mark) Disadvantage of using denaturing chemicals (1Mark) Membrane used (1Mark)	**07**
Q. 3 A	Give working principle of qRT PCR. Principle -2M Steps involved- 3M Analysis of result- 1M Diagram-1M	**08**
Q. 3 B	Discuss any three components of PCR. Principle of PCR-1M Any three component: 2M each • Template • Primer • DNA polymerase • Buffer systems used/master mix	**07**

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	• Deoxyribonucleotide Bases • Thermocycler	
	OR	
Q. 3 C	What is the function of DNA polymerase in PCR? What are the different types of DNA polymerases used in PCR method? Function- 2M Types: 6M • *Taq* polymerase • *Tth* polymerase • Stoffel fragment • ThermoSequenase • T7 Sequenase	**08**
Q. 3 D	How to control contamination in PCR? Significance of contamination-1M Methods Physical method-2M Chemical method-2M Use of enzyme-2M	**07**
Q. 4 A	Write a note on uses of genetic testing. Introduction -1M Uses: 1M each • Diagnostic testing • Carrier testing • Predictive • Susceptibility genetic testing Conclusion – 1M	**08**
Q. 4 B	Write a note on HIV and clinical significance of viral load in the serum sample? Introduction- 1M HIV structure-2M Virology -2M Correlation of viral load with prognosis of the disease-2M	**07**
	OR	
Q. 4 C	How RFLP is used to detect sickle cell anaemia? Introduction about sickle cell anaemia (Mutation)-2M RFLP principle- 1M Detection of sickle cell anaemia using RFLP- 3M Diagram-1M	**08**
Q. 4 D	Write a note on clinical significance of *N.gonorrhoeae*? 1M for each point • Common sexually transmitted microbe • Transmission requires direct, usually sexual contact • It can be symptomatic or asymptomatic • Intrapartum transmission can also occur, producing disease in neonates	**07**

	• Anogenital-Manifestations of disease include copious purulent rectal discharge, rectal pain, tenesmus, and bloody stools • Oropharyngeal infections- Mild pharyngitis • Both anogenital and oropharyngeal infections can be asymptomatic	
Q. 5	Write Short notes on any three of the following	**15**
a.	Northern blotting Significance- to investigate RNA structure, structural abnormalities and quantity (1Mark) Steps - RNase free environment, Agarose concentration, Denaturation, prehybridization and hybridization (4Marks)	
b.	Any one method for labelling of probe. End-labeling, nick translation, random priming (1 mark). (Any one method with a diagram) 4 marks	
c.	Reverse Transcriptase PCR Principle-1M Steps involved- 3M Diagram-1M	
d.	Mispriming Definition of misprimimg – 1M Reason for mispriming- 1M Prevention methods- 3M (any two) • Primer designing • Wax barrier • Hot start method	
e.	Any one commercial assays available to detect HIV1 virus. Introduction -1M Any one method- 4M	