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DEPARTMENT: MLT4TH

SUBJECT: MOLECULAR BIOLOGY

Q1: What is PCR? And explain the procedure and uses of PCR.

ANS:

POLMERASE CHAIN REACTION:

Polymerase chain reaction is a type of method in which millions and billions copy of a specific DNA is made.

PROCEDURE:

* PCR consist of a series of 20-40 repeated temperature changes, called cycle.
* Each cycle commonly consisting of 2-3 discrete temperature steps.
* Cycling is often preceded by a single temperature step called hold. At a high temperature >90 Degree c followed by one hold at the end for final product extension or brief storage.
* Temperature and length of time applied in each cycle depend on a variety of temperature.
* Enzymes used for DNA synthesis.
* Concentration of divalent ions.
* DNTPS in the reaction.
* Melting temperature of the primers.

USES:

* Gene cloning
* DNA sequencing
* DNA fingerprinting
* Genetically inherited diseases

Q2: Explain the process of AGAROSE gel electrophoresis.

ANS:

INTRODUCTION:

* It is a method which is used to separate RNA or DNA by size.
* It is achieved by moving negatively charged nucleic acid molecules through AGAROSE matrix with electric field.
* Shorter molecules moves faster than longer

METHOD FOR ELECTROPHORESIS:

* Prepare AGAROSE gel melt, cool and add ETHIDIUM bromide and mix thoroughly.
* Pour into casting tray with comb and allow to solidify
* And running buffer, load samples and marker.
* Run gel at constant voltage until band separation occurs.
* View DNA on UV light box and show result.

MATERIAL REQUIRED:

* Electrophoresis chamber
* AGAROSE gel
* Gel casting tray
* Buffer
* Staining agent
* A comb
* DNA ladder
* Sample to be separate