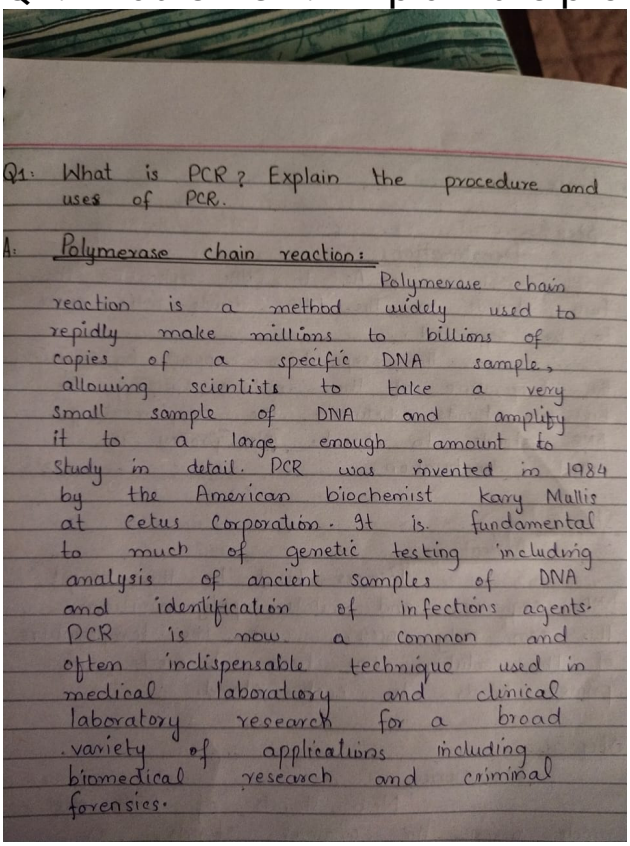
Assignment for viva

Q1.What is PCR? Explain the procedure and uses of PCR.



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

A: Polymerase chain reaction:

Polymerase chain reaction is a method widely used to rapidly make millions to billions of copies of a specific DNA sample, allowing scientists to take a very small sample of DNA and amplify it to a large enough amount to study in detail. PCR was invented in 1984 by the American biochemist Kary Mullis at Cetus Corporation. It is fundamental to much of genetic testing including analysis of ancient samples of DNA and identification of infections agents. PCR is now a common and often indispensable technique used in medical laboratory and clinical laboratory research for a broad variety of applications including biomedical research and criminal forensics.

→ Procedure:

Step 1:

Denaturation:

As in DNA replication, the two strands in the DNA double helix need to be separated.

The separation happens by raising the temperature of the mixture, causing the hydrogen bonds between the complementary DNA stands to break. This process is called denaturation.

Step 2:

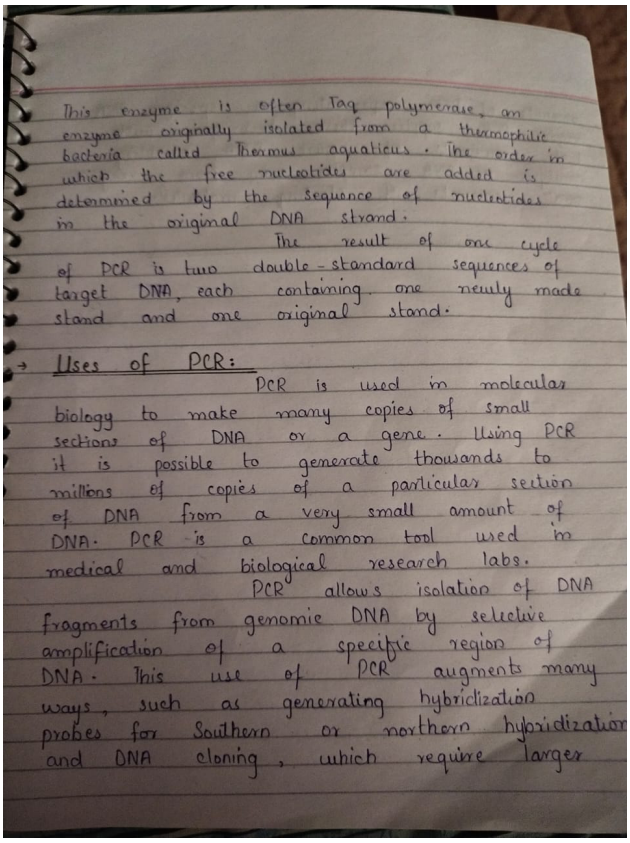
Annealing:

Primers bind to the target DNA sequences and initate polymerisation. This can only occur once the temperature of the solution has been lowered. One primer binds to each stand.

Step 3:

Extension:

New stands of DNA are made using the original stands as templates. A DNA polymerase enzyme joins free DNA nucleotides together.



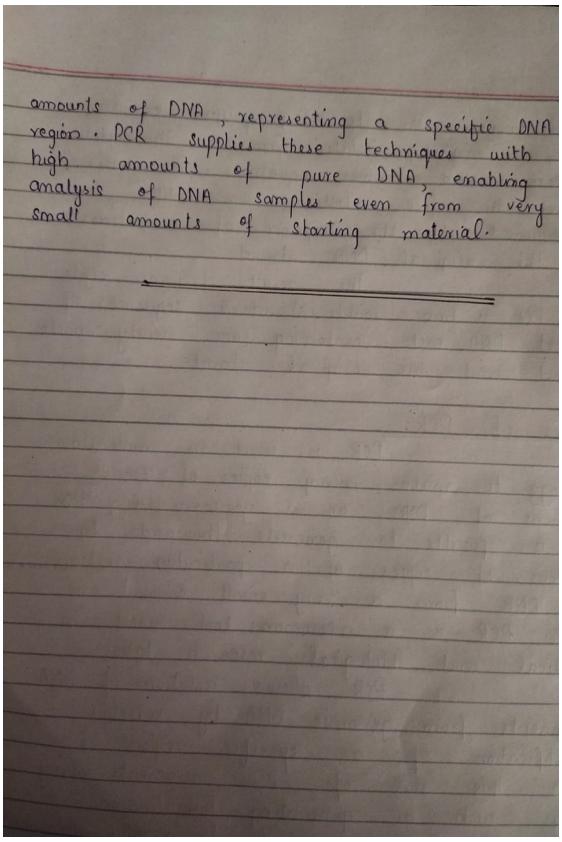
This enzyme is often Taq polymerase, an enzyme originally isolated from a thermophilic bacteria called Thermus aquaticus. The order in which the free nucleotides are added is determined by the sequence of nucleotides in the original DNA strand.

The result of one cycle of PCR is two double-standard sequences of target DNA, each containing one newly made stand and one original stand.

→ Uses of PCR:

PCR is used in molecular biology to make many copies of small sections of DNA or a gene. Using PCR it is possible to generate thousands to millions of copies of a particular section of DNA from a very small amount of DNA. PCR is a common tool used in medical and biological research labs.

PCR allows isolation of DNA fragments from genomic DNA by selective amplification of a specific region of DNA. This use of PCR augments many ways, such as generating hybridization probes for Southern or northern hybridization and DNA cloning, which require larger


amounts of DNA, representing a specific DNA region. PCR supplies these techniques with high amounts of pure DNA, enabling analysis of DNA samples even from very small amounts of starting material.

Q2.Explain the process of agarose gel electrophoresis

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A: Process of agarose gel electrophoresis:

The process of gel electrophoresis works because negatively charged molecules move away from the negative pole of the electronic current and smaller molecules will move faster than larger molecules.

Thus, a size seperation is achieved within the pool of molecules running through the gel.

The gel works in a similar manner to a sieve separating particles by size.

The electrophorsis works to move the particles, using their inherent electric charge, through the sieve.

When researchers are trying to distinguish between different sagments of DNA, for example, the process is simple.

The samples are loaded into channels at the start of the gel.

Each DNA molecule will have the same ford pulling it through the gel.

However, the size of each molecule hinders its progress through the gel. Large molecules hit parts of the gel matrix, and quickly make their way to the other side of the gel.

After a certain amount of time, the dyed DNA molecules can be seen aggregating in different areas of the gel, based on how far they moved during gel electrophoresis.

This allows researchers to identify the sagments, and compare the DNA of different organisms.