

Applications of Polymerase Chain Reaction

The top six applications are: (1) PCR in Clinical Diagnosis (2) PCR in DNA Sequencing (3) PCR in Gene Manipulation and Expression Studies (4) PCR in Comparative Studies of Genomes (5) PCR in Forensic Medicine and (6) PCR in Comparison with Gene Cloning.

Application # 1. PCR in Clinical Diagnosis:

The specificity and sensitivity of PCR is highly useful for the diagnosis of various diseases in humans. These include diagnosis of inherited disorders (genetic diseases), viral diseases, bacterial diseases etc. The occurrence of genetic diseases frequently identified by restriction fragment length polymorphism (RFLP) can be employed only when there is a mutation resulting in a detectable change in the length of restriction fragment. Many genetic diseases occur without the involvement of RFLP. For all such disorders, PCR technique is a real boon, as it provides direct information of DNA. This is done by amplification of DNA of the relevant region, followed by the direct analysis of PCR products.

Prenatal diagnosis of inherited diseases:

PCR is employed in the prenatal diagnosis of inherited diseases by using chorionic villus samples or cells from amniocentesis. Thus, diseases like sickle-cell anemia, p-thalassemia and phenylketonuria can be detected by PCR in these samples.

Diagnosis of retroviral infections:

PCR from cDNA is a valuable tool for diagnosis and monitoring of retroviral infections, e.g., HIV infection.

PCR is used for the detection of bacterial infection e.g., tuberculosis by *Mycobacterium tuberculosis*.

Diagnosis of cancers:

Several virally-induced cancers (e.g., cervical cancer caused by human papilloma virus) can be detected by PCR. Further, some cancers which occur due to chromosomal translocation (chromosome 14 and 18 in follicular lymphoma) involving known genes are identified by PCR.

PCR in sex determination of embryos:

Sex of human and livestock embryos fertilized in vitro, can be determined by PCR, by using primers and DNA probes specific for sex chromosomes. Further, this technique is also useful to detect sex – linked disorders in fertilized embryos.

Application # 2. PCR in DNA Sequencing:

As the PCR technique is much simpler and quicker to amplify the DNA, it is conveniently used for sequencing. For this purpose, single-strands of DNA are required. In asymmetric PCR,

preferential amplification of a single-strand is carried out. In another method, strand removal can be achieved by digesting one strand (usually done by exonuclease by its action on 5'-phosphorylated strand).

Application # 3. PCR in Gene Manipulation and Expression Studies:

The advantage with PCR is that the primers need not have complementary sequences for the target DNA. Therefore, the sequence of nucleotides in a piece of the gene (target DNA) can be manipulated and amplified by PCR.

By using this method, coding sequence can be altered (thereby changing amino acids) to synthesize protein of interest. Further, gene manipulations are important in understanding the effects of promoters, initiators etc., in gene expression.

PCR is important in the study of mRNAs, the products of gene expression. This is carried out by reverse transcription — PCR.

Application # 4. PCR in Comparative Studies of Genomes:

The differences in the genomes of two organisms can be measured by PCR with random primers. The products are separated by electrophoresis for comparative identification. Two genomes from closely related organisms are expected to yield more similar bands. For more details, refer the technique random amplified polymorphic DNA.

PCR is very important in the study evolutionary biology, more specifically referred to as phylogenetic. As a technique which can amplify even minute quantities of DNA from any source (hair, mummified tissues, bone, or any fossilized material), PCR has revolutionized the studies in palaeontology and archaeology. The movie 'Jurassic Park' has created public awareness of the potential applications of PCR!

Application # 5. PCR in Forensic Medicine:

A single molecule of DNA from any source (blood stains, hair, semen etc.) of an individual is adequate for amplification by PCR. Thus, PCR is very important for identification of criminals.

The reader may refer DNA finger printing technique described elsewhere.

Application # 6. PCR in Comparison with Gene Cloning:

PCR has several advantages over the traditional gene cloning techniques .These include better efficiency, minute quantities of starting material (DNA), cost-effectiveness, minimal technical skill, time factor etc. In due course of time, PCR may take over most of the applications of gene cloning.