

Role of Polymerase Chain Reaction in Clinical Medicine and Methodologies

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DESCRIPTION

The Polymerase Chain Reaction (PCR) is a revolutionary technique that has transformed the field of molecular biology. Developed in the 1980s by Kary Mullis, PCR allows scientists to amplify specific segments of DNA with remarkable speed and accuracy. This powerful tool has become essential for various applications in research, clinical diagnostics, forensics and biotechnology. This article discusses about the principles of PCR, its methodology, applications and future directions.

Principles of PCR

PCR consists of three main steps: Denaturation, annealing, and extension.

Denaturation: The reaction mixture is heated to around 94°C-98°C, causing the double-stranded DNA to separate into two single strands.

Annealing: The temperature is then lowered to about 50°C-65°C, allowing the primers to bind, or anneal, to their complementary sequences on the single-stranded DNA.

Extension: Finally, the temperature is raised to around 72°C, the optimal temperature for the enzyme Taq polymerase, which synthesizes new DNA strands by adding nucleotides to the primers. This cycle is repeated, typically 25 to 40 times, resulting in the exponential amplification of the target DNA sequence.

Methodology of PCR

PCR process requires specific components:

DNA template: This is the DNA sample containing the target sequence to be amplified. The DNA template serves as the starting material for PCR, providing the specific sequence that will be copied during the amplification process.

Primers: Short sequences that flank the target region, designed to ensure specificity in amplification. Primers bind to the complementary sequences on the DNA template, providing a starting point for Taq polymerase to initiate DNA synthesis.

Taq polymerase: A thermostable DNA polymerase derived from the bacterium *Thermus aquaticus*, capable of withstanding the high temperatures used during PCR.

Nucleotides: The building blocks of DNA, necessary for the synthesis of new DNA strands. Nucleotides serve as the fundamental units that make up DNA, consisting of a sugar, a phosphate group and a nitrogenous base.

Applications of PCR

The versatility of PCR has led to its widespread adoption across various fields.

Clinical diagnostics: PCR is invaluable in clinical settings for diagnosing infectious diseases. By amplifying pathogen DNA or RNA, clinicians can detect the presence of viruses and bacteria with high sensitivity and specificity. For instance, PCR testing has played a important role in the rapid detection of SARS-CoV-2, the virus responsible for COVID-19.

Forensic science: In forensic science, PCR is used to amplify DNA from crime scene samples, such as blood, hair or saliva. This allows investigators to generate profiles that can be compared to suspects or databases, significantly enhancing the ability to solve crimes.

Genetic research: PCR is a fundamental of genetic research, enabling scientists to amplify specific genes for further study, including sequencing, cloning, and mutational analysis. It facilitates the exploration of genetic variations associated with diseases and traits.

Biotechnology: In biotechnology, PCR is employed in the production of recombinant proteins, gene therapy and the development of Genetically Modified Organisms (GMOs). It enables precise manipulation of DNA, operating innovations in agriculture and medicine.

Future directions

As technology advances, PCR continues to evolve. Innovations such as quantitative PCR (qPCR) allow for the real-time monitoring of DNA amplification, providing quantitative data

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Received: 16-Aug-2024, Manuscript No. JMPB-24-34189; **Editor assigned:** 19-Aug-2024, PreQC No. JMPB-24-34189 (PQ); **Reviewed:** 02-Sep-2024, QC No. JMPB-24-34189; **Revised:** 09-Sep-2024, Manuscript No. JMPB-24-34189 (R); **Published:** 16-Sep-2024, DOI: 10.35248/jmpb.24.5.195

Citation: Tasleem S (2024). Role of Polymerase Chain Reaction in Clinical Medicine and Methodologies. J Mol Pathol Biochem. 5:195.

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about gene expression and pathogen load. Additionally, digital PCR (dPCR) offers enhanced sensitivity and accuracy by partitioning the PCR reaction into thousands of individual reactions.

Moreover, improvements in primer design and the development of new polymerases are expanding the capabilities of PCR, enabling amplification of challenging targets, including those with high Gas Chromatography (GC) content or secondary structures.

CONCLUSION

The PCR is a transformative technology that has revolutionized molecular biology and its applications. By enabling the rapid

and specific amplification of DNA, PCR has become indispensable in clinical diagnostics, forensic science, genetic research, and biotechnology. As the field continues to innovate, PCR will undoubtedly play a pivotal role in advancing our understanding of genetics and enhancing various scientific and medical disciplines, ultimately improving health outcomes and contributing to scientific discoveries.