

## Introduction

Polymerase Chain Reaction (PCR) is one of the most powerful and widely used techniques in molecular biology. Developed in 1983 by Dr. Kary Mullis, PCR allows scientists to amplify and replicate specific segments of DNA with remarkable precision and speed. This method has had a profound impact on fields ranging from genetics and diagnostics to forensics and medicine [1], enabling breakthroughs such as the identification of genetic diseases, DNA fingerprinting, and even the analysis of ancient biological samples. By rapidly creating millions of copies of a particular DNA sequence, PCR has revolutionized our ability to study and manipulate genetic material.

### The Basics of PCR

PCR is a laboratory technique used to create multiple copies of a specific DNA region. The process mimics natural DNA replication [2] but is carried out in a controlled environment outside of living cells. The basic components required for PCR include:

**DNA template:** The sample containing the DNA sequence to be amplified.

**Primers:** Short single-stranded DNA sequences that are complementary to the regions flanking the target DNA sequence. Primers are necessary to initiate the amplification process.

**DNA polymerase:** An enzyme that synthesizes new strands of DNA by adding nucleotides to the growing DNA chain. The enzyme used in PCR is typically Taq polymerase [3], derived from the thermophilic bacterium

environmental DNA (eDNA) for biodiversity studies, and monitor the health of ecosystems by detecting microbial DNA from soil or water samples.

### Advantages and Limitations of PCR

PCR offers numerous advantages, but it is not without its limitations.

#### Advantages

**Sensitivity:** PCR can amplify DNA from minute quantities of material, making it highly sensitive.

**Speed:** PCR can generate large amounts of DNA in a short time, usually within a few hours.

**Specificity:** The use of primers allows for the amplification of specific DNA sequences, minimizing the chance of non-specific amplification [9].

**Versatility:** PCR can be applied to a wide range of DNA sources, including ancient, degraded, or contaminated samples.

#### Limitations

**Contamination risk:** Because PCR amplifies DNA from very small amounts of starting material, contamination from external sources can lead to false results. Strict protocols are required to avoid contamination.

**Error rate:** While Taq polymerase (Taq pol) has a relatively low error rate, it is not perfect. The error rate is approximately 1 in 10<sup>4</sup> to 1 in 10<sup>5</sup> nucleotides. This can lead to mutations in the amplified DNA, which can affect downstream applications such as sequencing and cloning.