SCIE 100 PCR Write

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Polymerase Chain Reaction (PCR)

Polymerase Chain Reaction or PCR was introduced by Doctor Kary Mullis and his colleagues at the Cetus Corporation in 1986 (Mullis et al. 1986). In 1993, Doctor Kary Mullis and Professor Michael Smith received the Nobel Prize in Chemistry for the development of Polymerase Chain Reaction (NobelPrize 1993).

It is a thermal cycling process that uses different temperatures to duplicate a region of DNA to produce numerous copies of this particular region in DNA sequence within a short time period (Butler 2010). There are three main stages that occur in one cycle: denaturation, annealing, and extension (Gupta 2019). In the first stage, denaturation uses heat at 94°C to separate the double-stranded DNA into single-stranded DNA (Kubista et al. 2006). In the second stage, the temperature decreases from 95°C to 50°C to give primers the ability to bind to a single-stranded DNA (Kubista et al. 2006). In the third stage, the temperature increases from 50°C to 72°C to enable DNA polymerase to create a new copy of DNA by extending the primers using the deoxynucleotide triphosphates (Kubista et al. 2006).

PCR Cocktail is a combination of the components and reagents used to prepare an amplification reaction (Butler 2010). It may vary in ingredients from one PCR reaction to another PCR reaction, but it is mainly composed of buffer containing magnesium, deoxynucleotide triphosphates (dNTPs), forward and reverse primers, DNA polymerase, bovine serum albumin, and DNA template (Butler 2010). The most common DNA polymerase used in PCR is *Thermus aquaticus* or *Taq* because it can function at high temperature during the main stages (Innis et al. 1988). Deionized water is then added to the PCR cocktail to ensure that the desired concentration and volume of each of the components and reagents have been reached (Butler 2010). Once the PCR cocktail has been created, the samples can go into a thermocycler to perform the reaction.

Problems can arise during the amplification reaction such as contamination and inhibition (Andreas 2019). Controls are used to monitor the reaction (Butler 2010). Negative control is composed of PCR cocktail without DNA template (Butler 2010). If there is evidence of amplification in the negative control, it suggests that contamination occurred during the process (Butler 2010). Positive control is composed of PCR cocktail with a known DNA template (Butler 2010). It can determine if one or more of the components and reagents have failed to amplify during the reaction (Butler 2010). If there were no results in any of the samples, it indicates the samples that contains these components and reagents have failed to amplify (Butler 2010). These controls can determine the reliability of the results for the scientists.

Polymerase Chain Reaction (PCR) is the most common and widely used method in various fields such as Biology, Genetics, and Forensics (Shampo and Kyle 2002). It can help scientists learn and understand the living organisms including their genes.

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