UNIVERSITI TEKNOLOGI MARA

EFFECT OF VARIABLE ANNEALING TEMPERATURES IN POLYMERASE CHAIN REACTION

MOHD KHAIRUL SHAFIQ BIN ALI

Project submitted in fulfillment of the requirement for the Degree of Bachelor of Science (Hons.) Medical Technology.

FACULTY OF HEALTH SCIENCES

April 2007

This Final Year Project Report entitled **"Effect of Variable Annealing Temperatures in Polymerase Chain Reaction"** was submitted by Mohd Khairul Shafiq Bin Ali, in partial fulfillment of the requirement for the Degree of Bachelor of Science (Hons.) Medical Technology in the Faculty of Health Science, and was approved by

Dr. Zuridah Hassan
Supervisor
Faculty of Health Sciences
Universiti Teknologi MARA

En. Zed Zakari
Head of Programme
BSc. (Hons.) Medical Technology
Universiti Teknologi MARA

Prof. Dr. Abdul Rahim Md. Noor
Dean
Faculty of Health Sciences
Universiti Technologi MARA

Date: _____

ABSTRACT

EFFECT OF VARIABLE ANNEALING TEMPERATURES IN POLYMERASE CHAIN REACTION

Polymerase Chain Reaction (PCR) is typically carried out in three distinct steps governed by three distinct temperatures. These three important steps are the denaturing, annealing and extension. Annealing is the second stage of a PCR run whereby the primers will bind to the target DNA or target sequence for amplification. The goal is not to amplify *in vitro* the entire DNA but only specific region of interest only. In this study, the effects of variable or different annealing temperature on PCR performance were investigated. Six different annealing temperature setting were chosen, that is, 46^0C, 49^0C, 52^0C, 55^0C, 58^0C, and 61^0C. At the end of the PCR run, the amplicons were then electrophoresed, stained with ethidium bromide, destained in water and finally photographed by a gel documentation system with UV light. At 46^0C to 58^0C, DNA bands of varying intensity were produced. At 61^0C, no band was seen. These results showed that the best annealing temperature (Tm) for this consensus primer was 52^0C. The band was clear, sharp and with the highest intensity at the expected 1400 bp molecular weight. This therefore concludes that the Tm of a particular primer sets (forward and reverse) can be experimentally determined to obtain the optimum result.

TABLE OF CONTENTS