BINDURA UNIVERSITY OF SCIENCE EDUCATION

BIOLOGICAL SCIENCES DEPARTMENT

HBScBioTec

ANALYTICAL BIOTECHNOLOGY (BTEC133) (2)

EXAMINATION
2 HOURS (100 MARKS)

INSTRUCTIONS TO CANDIDATES

Answer FOUR questions. You MUST answer QUESTION 1 (Section A) and any THREE questions from section B. Each question carries 25 MARKS. Where a question contains sub-divisions, the mark value of each sub-division is given in brackets. Illustrate your answer where appropriate with large clearly labelled diagrams. You should not spend more than thirty minutes on each question.

SECTION A (COMPULSORY)

1. You are provided with 10 DNA samples and required to set up a PCR reaction for the amplification of exon 4 of the CYP2B6 gene. Calculate the concentrations and volumes (i) to (v) required for each reaction component except H_20 and DNA, and then calculate the required volumes for the master mix (vi) to (xii). (10 marks)

Table 1: PCR set up for CYP2B6 exon 4 amplification

Reaction component	Initial concentration	Final concentration	Initial Volume (µl)	Master Mix Vol (µl)
PCR buffer	(i)	1X	1.5	(vi)
dNTPs	2.5 mM	(iv)	1.2	(vii)
Primer 1	(ii)	0.4 µM	0.6	(viii)
Primer 2	(iii)	0.4 µM	0.6	(ix)
MgCl2	25 mM	(v)	0.6	(x)
Taq polymerase			0.5	(xi)
H_2O			9	(xii)
DNA			1	
Total volume			15	

(b) Describe five factors to consider when designing primers for PCR. (10 marks)

(c) Explain functions of $MgCl_2$ and buffer in a PCR reaction. (5 marks)

SECTION B

2. Discuss applications of immunoprecipitation in proteomics.

3. (a) Describe Sanger DNA sequencing. (15 marks)
(b) Highlight the uses of sequencing in recombinant DNA technology. (10 marks)

4. Write short notes on any **FIVE** of the following:
(a) Functions of loading buffer in gel electrophoresis. (5 marks)
(b) Western blotting. (5 marks)
(c) Applications of radioisotopes in biotechnology. (5 marks)
(d) Beer Lambert's law. (5 marks)
(e) Analytical centrifugation. (5 marks)
(f) Fluorescence microscopy. (5 marks)

5. (a) Describe an enzyme linked immunosorbent assay (ELISA) for detection of immunoglobulin for human immunodeficiency virus (HIV). (15 marks)
(b) Explain factors affecting immunoassays. (10 marks)

6. Discuss use of quantitative PCR in COVID19 diagnosis.

END OF EXAMINATION QUESTION PAPER