Polymerase chain reaction (PCR) and its applications

CREDIT DISTRIBUTION, ELIGIBILITY AND PRE-REQUISITES OF THE COURSE

Course title & Code	Credits	Credit distribution of the course			Eligibility criteria	Pre-requisite of the course (if any)
		Lecture	Tutorial	Practical/ Practice		
Polymerase chain reaction (PCR) and its applications	2	NIL	NIL	2	Class XII	NIL

Learning Objectives:

Students of this course should be able to learn:

- Concept of PCR and different types of PCR.
- Principles of oligonucleotide (primer) synthesis and purification
- Designing of Primers for PCR.
- Hands-on setting up of PCR reaction and analysis of the amplified product.
- Purification of PCR product

Learning Outcomes:

At the end of this course, students should be able to learn and perform:

- PCR and its application for research.
- Designing of Primers for PCR and obtaining Primers as synthetic oligonucleotides of
- appropriate quality from commercial sources.
- Designing conditions for setting up a PCR and perform analysis of amplified DNA.
- Purification of PCR-amplified DNA using columns for downstream application.

Unit 1 Concept of PCR and different types of PCR with some applications (12 hours)

Principle of Polymerase Chain reaction and amplification process, use of thermocycler and other equipment required to perform PCR and analysis of amplified DNA, use of synthetic oligonucleotide as primers in PCR.

Practical:

1.1 Demonstration of thermocycler, setting of conditions for a PCR for different types of templates and understanding the reagents used in PCR.

1.2 Chemistry of oligonucleotide synthesis and purification techniques, designing the Primer sequence for PCR through the use of online free software. Handling and storage of Primers for long term use.

Unit 2: PCR reaction for amplification and analysis of the amplified product (32 hours)

Designing and Setting up PCR, understanding about different reaction components, use of different types of polymerases for different length of amplicons, concept of fidelity and processivity of polymerases.

Practical:

2.1 Demonstration of a PCR to explain all the steps required for setting up the reaction and analysis of the amplified product.

2.2 Designing and setting up a PCR individually to amplify a 500 bp product and analysis of the amplified product using agarose gel electrophoresis.

2.3. Designing and setting up a PCR individually to amplify a 1500 bp product and analysis of the amplified product using agarose gel electrophoresis.

Unit 3: Purification of PCR-amplified DNA and downstream applications (20 hours)

Purpose of purification of PCR product, methods for purification, downstream applications of PCR products.

Practical:

3.1 Purification of PCR products by spin column method

3.2 Agarose gel electrophoresis and visualization of the product after purification.

Essential/ Recommended Readings:

1. Sambrook J, Fritsch EF & Maniatis T. Molecular Cloning. A laboratory Manual. 3rd Edition. Cold Spring Harbor Laboratory Press. New York.
2. Ausubel FM, Brent R, Kingston RE, Moore DD, Seidman JG, Smith JA, Struhl K. Current Protocols in Molecular Biology. (eds.) John Wiley & Sons, Inc. New York.

Suggestive Reading:

1. Alberts, B.,Bray, D, Lewis, J., et al. The Molecular Biology of the Cell. Garland Publishing, New York.

Note: Examination scheme and mode shall be as prescribed by the Examination Branch, University of Delhi, from time to time.