

WOLLO UNIVERSITY
DEPARTMENT OF BIOTECHNOLOGY
TECHNIQUES IN MOLECULAR BIOLOGY

DAY 1

1. introduction
 - 1.1. Laboratory safety rules
 - 1.1.1. Protective equipment
 - 1.1.2. Handling common laboratory equipment, chemicals, reagents, and other materials
 - 1.1.3. Laboratory organization
 - 1.2. Introduction to basic biotechnological laboratory equipment
 - 1.3. Laboratory report writing methods

DAY 2

2. Sterilization of equipment and other materials

- 2.1. chemical method
- 2.2. dry heat method
- 2.3. moist heat method

3. Solution preparations

- 3.1. stock solution preparation
- 3.2. working solution preparation
 - x Solutions preparation
 - Molar solutions preparation
 - normal solutions preparation

DAY 3

4. DNA isolation

- 4.1. Bacterial genomic DNA isolation
- 4.2. plasmid DNA
- Eukaryotic DNA isolation
- 4.3. DNA isolation from animal tissue.
- DNA purity determination by using Spectrophotometer

DAY 4

5. Restriction digestion of DNA

- 5.1. Single digestion
- 5.2. Double digestion
- 5.3. Triple digestion
- 6. PCR amplification of the desired gene
 - 6.1. principle of PCR
 - 6.2. preparation of reaction mixture
 - 6.3. PCR technique

DAY 5

7. Agarose gel electrophoresis

- 7.1. preparation of agarose gel and electrophoresis buffer
- 7.2. observing the nucleic acid bands in the gel documentation system
- 7.3. Elution of the DNA fragment from the gel

DAY 6

8. Cloning of foreign DNA fragment

- 8.1 Transformation of cells
 - Competent cell preparation
 - Calcium chloride method
- 8.2 screening of clones
 - blue white screening method
 - . antibiotic resistance method
- recombinant protein purification

DAY 7

- 8.3 Cell lysis
 - 8.3.1 physical means
 - 8.3.2 Sonication
- 9. Purification of expressed protein by chromatography
 - size exclusion chromatography
 - ion exchange
 - affinity chromatography

-Protein concentration determination

9.1. Bradford assay

DAY 8

10. Hybridization techniques

10.1. Colony hybridization

10.2. Southern blotting techniques

10.3. Northern blotting techniques

10.4. Western blotting techniques

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