Human Genetic

Lap 2

Instrument and material used in genetic lab

By

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Human Genetics

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Lab 2

Instrument and material used in genetic lab

Genetics: is the branch of biology that deals with the structures of genetic material and the transmission of characters from one generation to another.

Accreditation Requirements for Molecular Genetics Laboratories

1. Requisitions and Specimen Receipt

All specimens should be accompanied by a requisition form which contains as much of the following information as possible: unique patient identification, sex, date and time of specimen collection, specimen type, race/ethnicity, unique identifier found on the specimen container, tests requested, patient location, reason for requesting the test, relevant clinical or laboratory information, pedigree (required for linkage analysis, recommended for all cases), referring physician or health professional and billing information.

2. Specimen Processing

Sample identification should be assured through all applicable phases of analysis including nucleic acid extraction and quantification, restriction enzyme digest, electrophoresis, transfer, hybridization, detection, in situ hybridization, enzymatic amplification, photography and storage. Sources for DNA isolation are very diverse. Basically, it can be isolated from any living or dead organism. Common sources for DNA isolation include whole blood, hair, sperm, bones, nails, tissues, blood stains,

saliva, buccal (cheek) swabs, epithelial cells, urine, paper cards used for sample collection, bacteria, animal tissues, or plants.

3- Nucleic Acids Extraction

The quantity of nucleic acid should be measured and recorded. This is usually done using a spectrophotometer that has been properly calibrated with the use of proper controls and measuring the absorbance. This should be performed in clean, dry, quartz cuvettes within the linear range of the particular spectrophotometer being used. To determine the concentration of purified DNA, an absorbency reading at 260 nm (nucleic acid absorbs maximally at this wavelength) should be performed. Since proteins absorbs maximally at 280 nm, determination of the A260/A280 ratio provides a qualitative measurement of the level of DNA in respect to the amount of contaminating protein in the Sample, Long-term storage should be carried out at -20°C or -70°C to Long-term storage should be carried out at -20°C or -70°C to prevent degradation. RNA should be prevented degradation. RNA should be stored stored at -20°C -20°C or -70°C once -70°C once extracted, extracted, since RNA degrades quickly.

4- Addition of Reaction Components Non-sample components (mineral oil, dNTPs, primers, buffer and enzymes) should be added to the amplification reactions before addition of the sample.

Instrument and material used in genetic lab:

Refrigerator

The device is used for storage of stock solutions, chemical, kit, and PCR products that should be maintained at certain temperature.

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Deep freezer

*It is used to store mammalian cell stock culture, it is a device used to store materials when should be kept at low temperature (cell, tissues, enzyme, protein, etc.)
*This instrument is defined as freezers for -80 to -85°C and the inner volume inside are in general between 300 and 800 L.
* **Uses:** for long term storage for biological samples like DNA, RNA, proteins, cell extracts, or reagents. To reduce the risk of sample damage, these types of samples need extremely low temperatures as -80 to -85°C.

Refrigerator (Beko)

Deep freezer

Magnetic stirrer.

Is advice used which proved mixing and keeping chemical solution at a certain time and temperature by the help of magnetic bar.

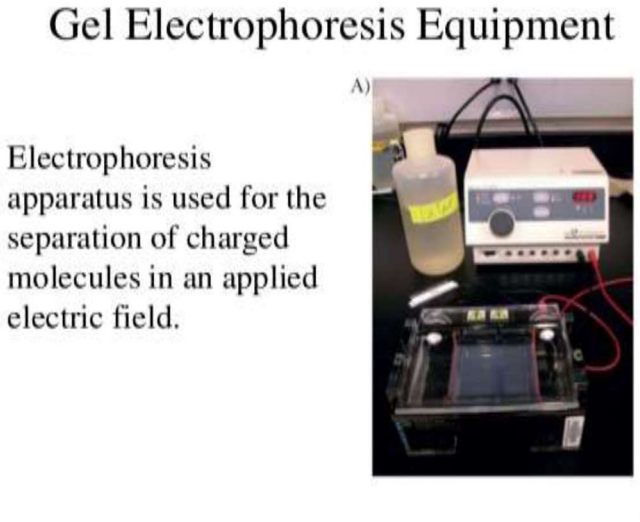
vortex

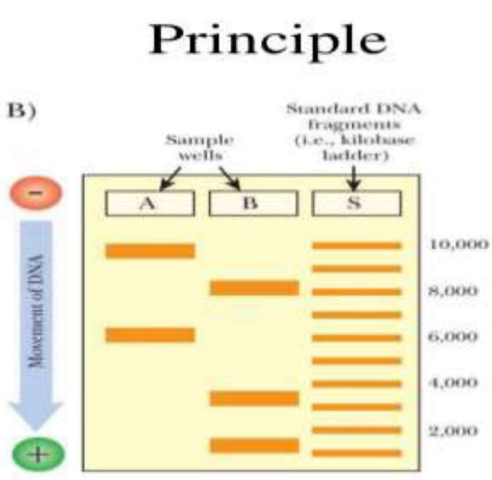
*It consists of an electric motor and attached to a cupped rubber piece.

* It used to mix sample at certain speed and duration, is a simple device used commonly in laboratories to mix small vials of liquid.

*When a test tube or other appropriate container is pressed into the rubber cup (or touched to its edge) the motion is transmitted to the liquid inside test tube or another appropriate container and a vortex is created.

Gel Electrophoresis





1. Major components

- Box to hold the gel
- Comb to create small wells in the agarose gel to put the DNA sample into at the beginning of the gel Positive and negative electrodes to create the electrical current
- Power supply
- Gel photo imaging system

Applications of Gel Electrophoresis

DNA can be separated by electrophoresis to

1- Visualize bands of a molecular marker to genotype individual
2- Verify amplification by PCR or sequencing reactions
3- Check the quality and quantity of genomic DNA after DNA extraction
4- Used for investigating the DNA
5- To investigate various binding modes of small molecules to supercoiled DNA
6- Complex mixtures can be separated to very high resolution by this process .

Staining of gel

One of the most important aspects of gel electrophoresis technique is staining. once sample molecules have separated in the gel matrix it is necessary to visualize their position. This is achieved by staining with an agent appropriate for the sample.

Stain	Used	Detection limited (ng)
Amino black	Protein	400
Coomassie blue	Protein	200
SYPRO red	Protein	0.5
Sliver chloride	Protein /DNA	1
Ethidium bromide	DNA /RNA	10

Gel documentation

This device used to display DNA fragment after electrophorese

PCR (thermal cycler or DNA amplifier):

This device is used for the amplification of specific region of any DNA sample with polymerase chain reaction in test tube. It is also used for detection and constituent of genetically modified organisms as well as other genetic analyses.

Thermal Cycler (Agilent)

***PCR is polymerase chain reaction.**

Principle of PCR: a small fragment of the DNA section of interest needs to be identified which serve as template for producing the primers that initiate the reaction.

Applications.

- Genome mapping and gene function determination
- Biodiversity studies (e.g. evolution studies)
- Diagnostics (prenatal testing of genetic diseases, early detection of cancer, viral infections, mutation detection ...)
- Detection of drug resistance genes
- Forensic (DNA fingerprinting) Microarrays , molecular cloning , recombinant DNA research .

pH Meter

Biological functions are very sensitive to changes in pH and hence, buffers are used to stabilize the pH. A pH meter is an instrument that measures the potential difference between a reference electrode and a glass electrode, often combined into one combination electrode.

pipette

(Sometimes spelled pipet) is a laboratory tool commonly used in chemistry, biology and medicine to transport a measured volume of liquid.

Pipette type	Volumes (µL)	Tip color
P 10	0.5-10	white
P20	2–20	yellow
P 200	20-200	yellow
P 1000	100-1000	blue

Micro centrifuges

is a piece of laboratory equipment, driven by a motor, which spins liquid samples at high speed, there are various types of centrifuges, depending on the size and the sample capacity laboratory centrifuges work by the sedimentation principle, where the centripetal acceleration is used to separate substances of greater and lesser density. devices for small tubes from 0.2 ml to 2.0 ml (micro tubes), up to 96 well-plates, compact design, small footprint; up to 30,000 g of ten used to isolate nucleic acids such as DNA.

Electronic balances: - use to quickly and accurately measure the mass of a substance to a level of accuracy impossible for traditional balances to achieve. This is especially important in experiments that require precise amounts of each substance to achieve the desired results.

Electronic balances

UV and Visible Spectroscopy

- UV - VIS spectroscopy is used to detect the presence of chromophores like dienes, aromatics, polyenes, and conjugated ketones, etc
- It uses light in the visible and adjacent ranges.
- The absorption or reflectance in the visible range directly affects the perceived color of the chemicals involved.
- This technique is complementary to fluorescence spectroscopy.