Section

I

Laboratory Instruments and Biosafety

Chapter

1

Commonly Used Laboratory Apparatus and Equipment

Competencies

BI 11.1: Describe commonly used laboratory apparatus and equipment.
BI 11.19: Outline the basic principles involved in the functioning of instruments commonly used in a biochemistry laboratory and their applications.
BI 11.16: Observe use of commonly used equipment/techniques in biochemistry.

Various instruments and equipment used in undergraduate biochemistry lab and clinical biochemistry lab are:

- Balances
- Magnetic stirrer
- Centrifuge
- Hot air oven
- Incubator
- Water bath
- Desiccator
- pH meter
- Thermometer
- Various glasswares
- Semiautoanalyzer
- Fully autoanalyzer
- Electrophoresis (described in Chapter 35)
- Chromatography (described in Chapter 36)
- Colorimeter/photometer/spectrophotometer (described in Chapter 5)

There are many instruments which are being used in different biochemistry lab for teaching and diagnostic purposes. Their description is given in this chapter along with the principle of their functioning and the application.

Balances

It is important to prepare reagents and standard solution with accurate measurement of various chemicals. For this purpose, the instrument used is "Balance".

There are various types of balances used in biochemistry lab. They are:

a. Physical/mechanical balance
b. Electronic balance

Physical (Mechanical) Balance

- It is rather a crude equipment which is used for measuring chemicals which are needed for making qualitative reagents and where larger amount of chemical need to be measured.
- They are double-pan balances where on one side known weight is kept and on other side chemical to be measured is equated.
- The weight which can be measured in this instrument vary from **1 mg to 1000 g**.
- Physical balance may be **open** two-pan balance or it may be a balance in **closed** compartment (Fig. 1.1).

Fig. 1.1: Physical balance in closed compartment

Electronic Balances

They are more accurate and are being preferred because of accuracy and ease of operation. They are **single pan balances** which operate on **principle of electromagnetic force** which counterbalance the weighed sample mass. It measures rather smaller amount compared to physical balance. The weight which can be measured ranges from **1 mg to 100 g**.

Facility of **"taring"** in electronic balances makes the measurement easier as because of it the weight of container is adjusted by the machine itself (Fig. 1.2).

Magnetic Stirrer

This is used for thorough mixing of solutes in the solution to make a homogenous mixture. An iron capsule is placed in the vessel containing solution and then this vessel is placed on the pan of magnetic stirrer plate which is built to provide rotating magnetic field.

Movement of iron capsule in the solution on switching on the instrument mixes the solution completely as to provide the homogenous mixture (Fig. 1.3).

Fig. 1.2: Electronic balance

Fig. 1.3: Magnetic stirrer

Centrifuge

This instrument is commonly used in biochemistry lab to separate serum or plasma from whole blood and it also separates sediments in the urine (Fig. 1.4).

Centrifuge is based on the **principle of centrifugal force** which is used to separate solid matters from liquid suspension.

Fig. 1.4: Centrifuge

Centrifuge can be classified as:

a. Bench-top or floor model
b. With refrigeration and without refrigeration
c. Based on rotor design
d. Based on maximum speed which can be attained (normal or ultracentrifuge)

Rotor is the component of the centrifuge machine which is holding the tubes or vacutainers. Based on rotor design, we have two types of centrifuge:

- Horizontal rotor type (swing out head)
- Fixed angle rotor type (angle head)

Fixed angle rotor type of centrifuge can be operated at high speed compare to horizontal rotor type.

This is the reason that lesser air friction is generated in fixed angle type resulting in lesser heat production. In swing out rotor type of centrifuge, more air friction results in more heat production, hence it cannot be operated at very high speed.

Principle of Centrifuge Machine

Key factor in separating the particulate matter from liquid suspension is the **relative centrifugal force (RCF)** which is represented by the following formula

$$\boxed{\text{RCF} = \text{R} \times (\text{RPM})^2 \times 118 \times 10^{-7}}$$

R is the rotating radius of the rotor which means the radius of rotating path from the central axis.

RPM (revolution per minute): It is the number of revolutions of rotor in a minute. It can be programmed in the centrifuge.

Ultracentrifuge: The centrifuge which can be operated at **very high speed (1,00,000 RPM)** is known as ultracentrifuge.

Hot Air Oven

It is the equipment which produces dry heat in its internal chamber, the temperature may go up to 250°C. These were originally developed by Pasteur. It has double-walled insulation which conserves the heat (Fig. 1.5).

This is used for:

1. Drying of the glassware
2. Heating the chemicals wherever required
3. Dry sterilization of glassware (in microbiological experiments), metal equipment, swabs, etc.

Fig. 1.5: Hot air oven

Incubator

It is an instrument which is capable of maintaining desired temperature, humidity, oxygen and CO_2 in the atmosphere inside (Fig. 1.6).

It is mainly used in microbiology for culture of organism under specified conditions. In biochemistry, incubator is important to incubate the reaction mixture at specific temperature for set duration which depends upon the method adopted for assessment.

Here the temperature may go up to 60–65°C, never beyond 100°C.

Fig. 1.6: Incubator

Water Bath

This equipment has water filled in it, the temperature of which can be controlled by adjustment (Fig. 1.7).

Fig. 1.7: Water bath

This is used for various experiments where a specified temperature is used for heating the reaction mixture. It is commonly used in medical undergraduate practicals where heating of solution is desired. In this equipment, sample can be heated at constant temperature over a prolonged period of time.

Desiccator and Desiccants

Desiccants are hygroscopic substances which absorb water from atmosphere and from various other hydrated substances. In other words, desiccants are **drying agents**. In biochemical lab, we need to remove moisture from many chemicals before using them for analytical purpose, for that purpose these chemicals are kept in contact with desiccants in a closed, airtight chamber which has a lid with a lubricated seal known as desiccator (Fig. 1.8).

Fig. 1.8: Desiccator

pH Meter

What is pH?

pH of a solution is negative logarithm of the hydrogen ion concentration. It can be expressed as:

$$pH = 1/\log 10\ (H^+)$$

As stated in equation above, pH of a solution is inversely proportional to its hydrogen ion concentration. Normal pH of some important biological fluids in human is enumerated below:

- Plasma = 7.35 to 7.45 [Mean = 7.4]
- Urine = 6.5
- CSF = 7.32
- Gastric juice = 1.0 to 2.0

pH meter: pH meter is a reliable and convenient method of measuring the pH of a solution. The pH meter measures the difference in electric potential between a pH electrode and a reference electrode, and so the pH meter is sometimes referred to as a **"potentiometric pH meter"** (Fig. 1.9).

Fig. 1.9: pH meter

Principle of its Functioning

The electrodes, or probes, are inserted into the solution to be tested. On immersion in the solution to be tested, hydrogen ions in the test solution exchange for other positively charged ions on the glass bulb, creating an electrochemical potential across the bulb. The electronic amplifier detects the difference in electrical potential between the two electrodes generated in the measurement and converts the potential difference to pH units.

This instrument has the following components in it:

- Glass electrode
- Calomel electrode

Glass electrode: It has a thin-walled bulb which is filled with 0.1 M HCl. In this HCl, a silver wire coated with AgCl is dipped (Fig. 1.10).

The hydrogen ion selective glass membrane has the following composition:

- CaO: 6%
- N_2O: 22%
- SiO_2: 72%

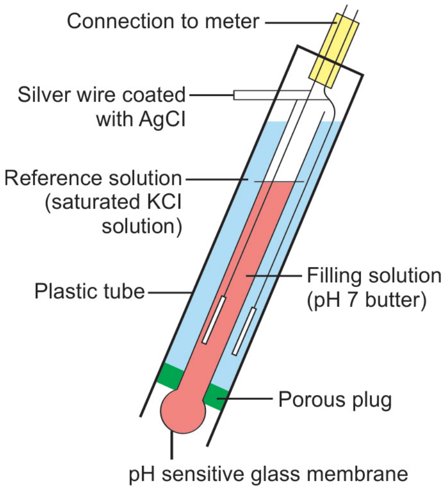


Fig. 1.10: pH meter (glass electrode)

Calomel electrode: It consists of glass tube which is filled with saturated solution of KCl and KCl porous material is plugged at the tip. In this solution, mercuric-mercurous electrode is dipped, the tip of which has porous plug of mercury calomel paste.

Combined electrode: In actual instrument, reference electrode and measuring electrodes are combined.

Thermometer

Many analytical reactions require an optimal temperature at which they occur. Enzymatic reactions specially need precise control of temperature. Many other reactions in biochemical system occur at wide range of temperature. In addition to this, many instruments like laboratory refrigerator, incubators, hot air oven, etc. need to have temperature display to monitor their accurate functioning.

Thermometer is the device which is used to monitor the temperature of the environment. It may be an integral part of the instrument or can be placed in the device for this purpose. Three major types of thermometers are:

1. Liquid in glass thermometer
2. Electronic thermometer (thermistor)
3. Digital thermometer

Liquid in glass thermometer has coloured liquid or mercury which is filled in glass or transparent plastic stem with bulb at one of its ends. Stem is graduated to record the temperature (Fig. 1.11).

In era of automation, electronic thermometer (thermistor) is incorporated in many devices which give accurate and fast reading.

Fig. 1.11: Thermometer

Glasswares

A number of glasswares are used in biochemistry laboratories. The glassware which is used to transfer the liquid may be the one which is to contain (TC) or to deliver (TD) type based on how accurately the specified volume of liquid is transferred.

Various glasswares are described below:

- Flask
- Griffin beaker

- Graduated cylinder
- Pipettes
- Burets

Flask

Volumetric flask: This flask has round lower portion with flat bottom, along with a long narrow neck where calibrated line is marked. This kind of flask is used to prepare a specific volume of reagent (Fig. 1.12).

Erlenmeyer flask: This flask has wide bottom with narrow neck and is designed to hold different volumes of liquid. For this purpose, there is marking of various volumes on its wall (Fig. 1.13).

Fig. 1.12: Volumetric flask

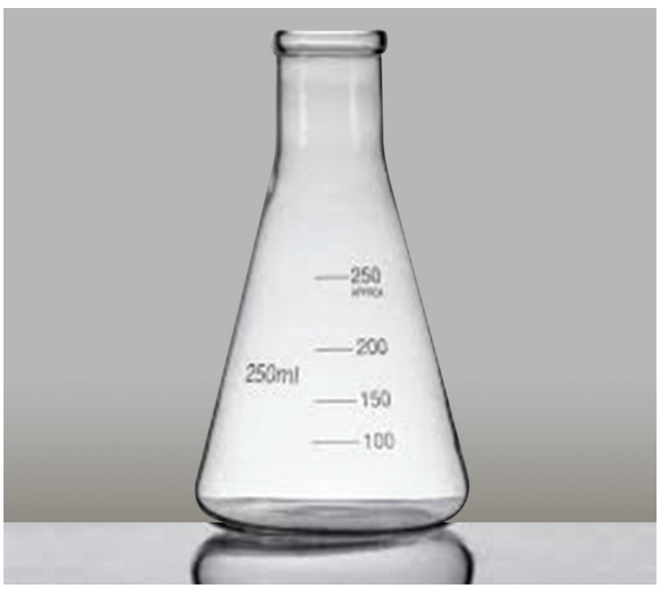


Fig. 1.13: Erlenmeyer flask

Griffin Beaker

It has flat bottom and straight wall with wide opening. It has marking on its side wall and can be used to store various volumes of solution (Fig. 1.14).

Fig. 1.14: Griffin beaker

Graduated Cylinder

These are cylinders having long stem and round or octagonal base. The side wall has markings of various volumes and this is used to measure volume of the reagent. It is available in various capacities from 10 to 2000 mL (Fig. 1.15).

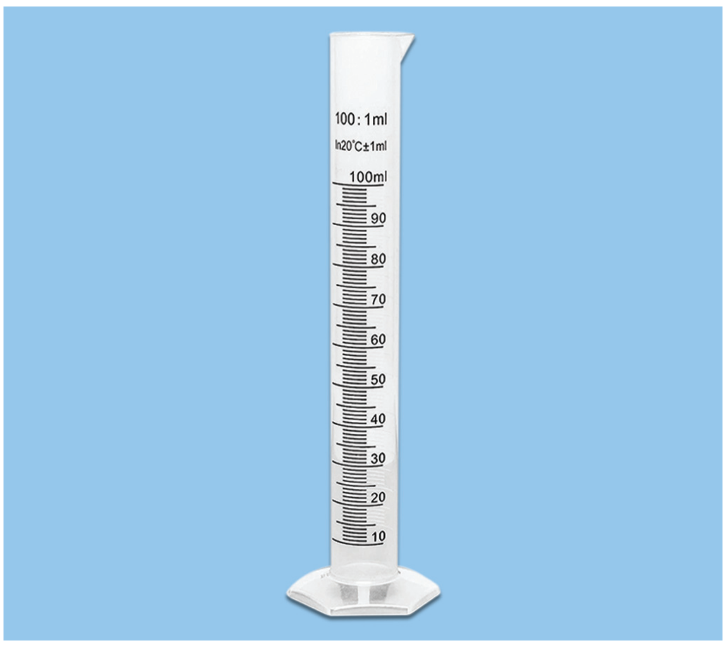


Fig. 1.15: Graduated cylinder

Pipette

Pipettes may be **TC (to contain) or TD (to deliver)** type depending upon whether it is delivering the exact specified volume or not as explained above.

Depending upon the draining characteristic, pipette may be **self-draining type or blowout type. Blowout pipette has got two continuous rings at the top.** Pipettes without this marking are self-draining type.

There are two major classes of pipettes: Measuring (graduated) and transfer types.

Measuring (graduated) pipettes: They are designed to dispense different volumes for which they have markings on it. They are of two types:

1. *Mohr type*: Mohr pipette does not have graduations till tip and it is self-draining type.
2. *Serological type*: Serological pipette has graduation till tip and is blowout type (Fig. 1.16).

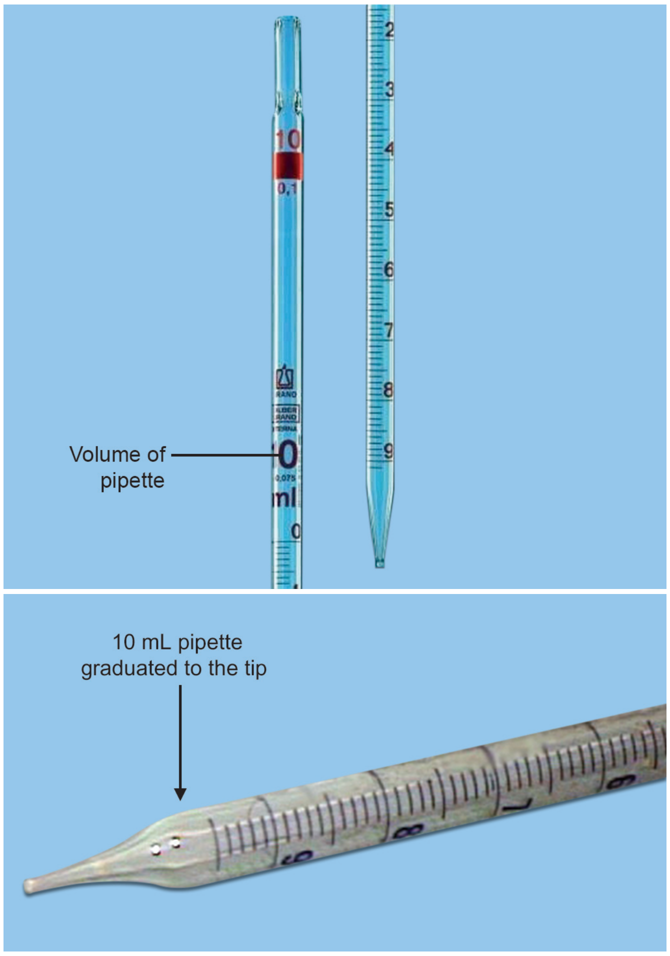


Fig. 1.16: Serological blowout pipette. Notice the marking till tip

Transfer type of pipettes: They are designed to deliver a specific volume alone. They are of following types (Fig. 1.17):

1. *Volumetric*: This type of pipette has bulb in it and it is self-draining type.
2. *Ostwald-Folin*: This type of pipette has bulb in it and is blowout type.
3. *Pasteur*: No calibration mark, not used for quantitative test (Fig. 1.18).

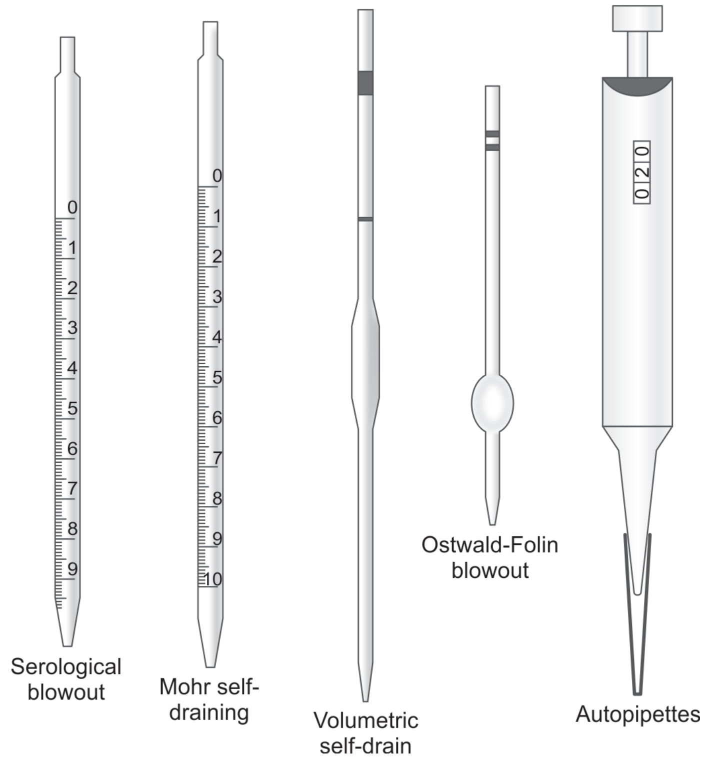


Fig. 1.17: Various types of pipettes

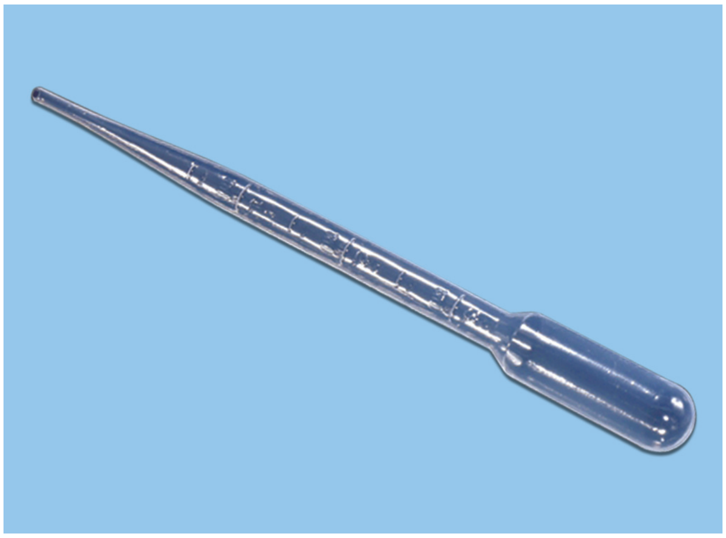


Fig. 1.18: Pasteur pipette

Nowadays **autopipettes** are used commonly to transfer volume less than 1 mL. They are easy to use and accuracy is high. They are of mainly two varieties: **Fixed volume and variable type.** Those which are designed to transfer volume of fluid less than 1 mL are called micropipettes and those which transfer larger volume of liquid are called **automatic macropipettes** (Fig. 1.19).

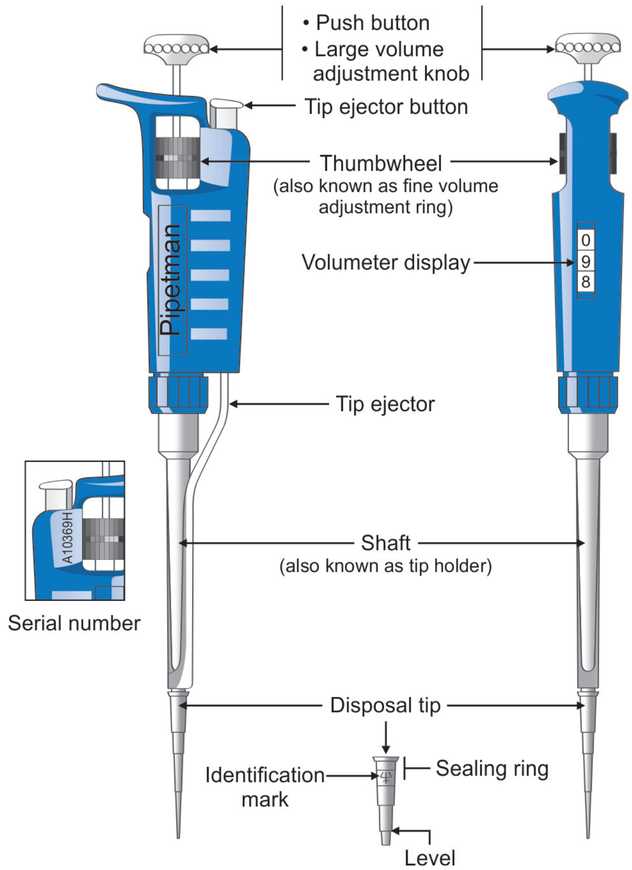


Fig. 1.19: Autopipette

Burets

It is long, graduated big pipette-like structure with a stopcock at the lower end. Its volume ranges from 25 to 100 mL. It is used to dispense desired volume of liquid drop by drop (Fig. 1.20).

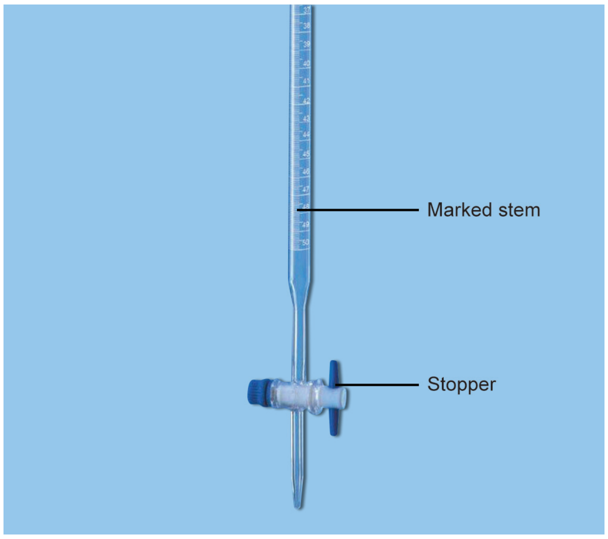


Fig. 1.20: Burets

Semiautoanalyzer

Principle and Functioning

Semiautomatic biochemistry analyzer is capable of performing routine biochemistry, hormonal assay, electrolytes, therapeutic drugs and drug-enzyme investigations. This instrument is based on principle of photometry described under colorimeter description in Chapter 5.

Semiautomated biochemistry analyzer is capable to perform tests on whole blood, serum, plasma, cerebrospinal fluid and urine as sample and it is suitable for small-sized laboratory (Fig. 1.21).

Fig. 1.21: Semiautoanalyzer

Fully Autoanalyzer

Principle and Functioning

The automatic biochemical analyzer has automated the sequence of operation processes which used to be conducted manually in past (Fig. 1.22).

It is based on colorimetric principle.

Important components of fully autoanalyzer are given in Fig. 1.23.

Fig. 1.22: Fully autoanalyzer

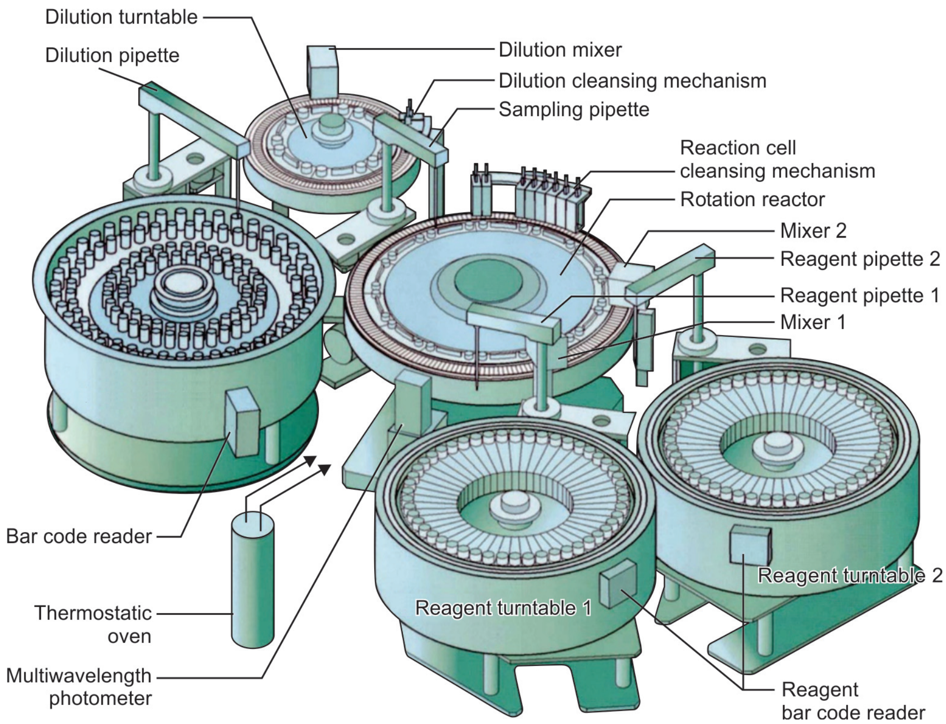


Fig. 1.23: Core components of fully automated biochemistry analyzer

Following are important parts in a fully autoanalyzer:

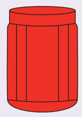
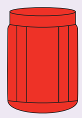
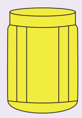
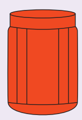
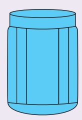
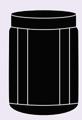
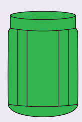
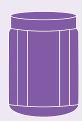
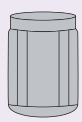
1. Reagent carousel
2. Sample compartment
3. Reaction cell
4. Sample probe (pipette)
5. Reagent probe (pipette)
6. Dilution pipette
7. Mixer

There are varieties of blood collection tube (vacutainers) available for collection of blood for specific purposes.

Their types, color of cap, additives (if any), purpose and mixing instruction is given in Table 1.1.

Fig. 1.24: Actual picture of various types of vacutainers

Table 1.1: Various types of vacutainers as coded by colour of its cap and their purpose

Type of vacutainer/ Colour of the cap	*Representation*	*Additive*	*Purpose*	*Mixing instruction*
Red [glass] [clotting time: 60 min]		Nil	Serum biochemistry	Do not invert
Red [plastic] [clotting time: 60 min]		Silicone coating	Serum biochemistry	Invert 4 times
Yellow [clotting time: 30 min]		Clot activator	Serum biochemistry	Invert 4 times
Orange [clotting time: 5 min]		Rapid serum tube [thrombin based clot activator]	Serum biochemistry	Invert 4 times
Light blue		Sodium citrate [1:9]	Coagulation studies	Invert 4 times
Black		Sodium citrate [1:4]	Coagulation study	
Green		Heparin	ABG analysis	Invert 8 times
Lavender/purple		EDTA	Hemogram, ESR, HbA1c	Invert 8 times
Gray		Potassium oxalate and sodium fluoride	Blood glucose, blood alcohol	Invert 8 times

VIVA VOCE

Q1. How many types of balances are being used in biochemistry lab?

Ans. There are various types of balances used in biochemistry lab. They are:

a. Physical/mechanical balance
b. Electronic balance

Q2. What do you understand by term "Taring" in electronic balance?

Ans. Facility of "taring" in electronic balances makes the measurement easier as because of it the wait of container is adjusted by the machine itself.

Q3. What is magnetic stirrer and why it is used?

Ans. This is used for thorough mixing of solutes in the solution to make a homogenous mixture

Q4. How many variations of centrifuge you know?

Ans. Centrifuge can be classified as

a. Bench-top or floor model
b. With refrigeration and without refrigeration
c. Based on rotor design
d. Based on maximum speed which can be attained (normal or ultracentrifuge)

Q5. What is the rotor in a centrifuge?

Ans. Rotor is the component of the centrifuge machine which is holding the tubes or vacutainers. Based on rotor design, we have two types of centrifuge:

- Horizontal rotor type (swing out head)
- Fixed angle rotor type (angle head)

Q6. What is the principle of centrifuge?

Ans. Principle of centrifuge machine

Key factor in separating the particulate matter from liquid suspension is the relative centrifugal force (RCF) which is represented by the following formula

$$RCF = R \times (RPM)^2 \times 118 \times 10^{-7}$$

R is the rotating radius of the rotor which means the radius of rotating path from the central axis.

RPM (revolution per minute): It is the number of revolution of rotor in a minute. It can be programmed in the centrifuge.

Q7. What is ultracentrifuge? Mention its use.

Ans. The centrifuge which can be operated at very high speed (1,00,000 RPM) is known as ultracentrifuge.

This is used for:

1. Drying of the glassware
2. Heating the chemicals wherever required
3. Dry sterilization of glassware (in microbiological experiments mainly), metal equipment, swabs, etc. (in surgical units).

Q8. What is desiccator and what is desiccant?

Ans. Desiccator and desiccants

Desiccants are hygroscopic substances which absorb water from atmosphere and from various other hydrated substances. In other words, desiccants are drying agents.

In biochemical lab, we need to remove moisture from many chemicals before using them for analytical purpose, for that these chemicals are kept in contact with desiccants in a closed, airtight chamber with a lid with a lubricated seal is known as desiccator.

Q9. What is pH?

Ans. pH of a solution is negative logarithm of the hydrogen ion concentration. It can be expressed as

$$pH = 1/\log 10\ (H^+)$$

As stated in equation above, pH of a solution is inversely proportional to its hydrogen ion concentration.

Plasma = 7.35 to 7.45 [Mean = 7.4]
Urine = 6.5
CSF = 7.32
Gastric juice = 1.0 to 2.0

Q10. Mention the difference between Mohr pipette and serological pipette.

Ans.

Mohr type: Mohr pipette does not have graduations till tip and it is self-draining type.

Serological type: Serological pipette has graduation till tip and is blowout type.

Notes

Chapter 2

Safe Laboratory Practice and Waste Disposal

Competency

BI 11.1: Describe good safe laboratory practice and waste disposal.

LABORATORY SAFETY

All clinical laboratory personnel are constantly exposed to various hazards like electric shock, radioactive material hazard, gaseous hazard, corrosive substances, and risk of handling biological material.

Recognition of hazard in the lab is of foremost importance along with the right attitude of the employer and employee towards safety measures to have a safe lab practice.

Safety Awareness for Laboratory Personnel

- Both employer and employee need to share the responsibility for effective implementation of safety practices in the lab.
- Employer need to formulate the laboratory work methods and safety policies and should provide time-to-time training to all employees.
- Personal protective equipment (PPE) consisting of gloves, eye shield, apron, cap, shoe cover should be provided to them. Employee should comply with the guidelines and should have positive attitude towards safety practices.
- Signage and labelling should be used liberally to identify the critical hazards and guidelines for precautions should be displayed at appropriate places.

Biological Safety

Clinical lab personnel deal with potential infectious blood and other biological samples. Utmost care should be taken while collecting, transporting, processing and analyzing such samples. Proper gloves, gowns, face protection should be practiced while handling such samples.

Any infectious material (blood, urine, or any other biological material), if spilled, should be taken care. 10% bleach should be used at spill site for appropriate time and then the site should be rinsed with water.

All the blood and other biological samples should be handled taking universal precaution considering all of them as potentially infectious.

Chemical Safety

Material safety data sheets (MSDS) for each hazardous compound at workplace should be obtained and all employees should be educated for its use and they should be clearly explained how to work safely with the chemicals.

Fire Safety

Fire extinguisher should be placed at appropriate place and all lab personnel should be trained for its use at the time of need.

Radiation Safety

All the places where radioactive material is stored should be labelled with cautious signage and entry of only authorized personnel should be allowed.

Disposal of Hazardous Material

Chemical material disposal: Most of the water-soluble chemicals may be flushed in the drain with large amount of water. Strong acid and strong bases should be neutralized first before discarding them in drain.

Possible chemical reaction in the drain between certain chemicals should be kept in mind and due precaution is to be taken. Foul smelling chemicals should not be disposed directly, rather they should be diluted first before being dumped in the drain.

Solid chemical waste can be buried in a landfill.

Radioactive material disposal: Radioactive material should be handed over to licensed receiver for safe disposal.

Handling Accidental Exposure to Acid and Alkali

Exposure to Acid

Various acids used in biochemistry laboratory are: Hydrochloric acid, nitric acid, sulfuric acid and acetic acid.

Exposure to skin

1. Wash with large quantity of water.
2. Apply 5% sodium carbonate solution with cotton wool.

Exposure to eye

1. Spray large quantity of water in eye.
2. Add 3 to 4 drops of 2% sodium carbonate in eye every 5 minutes till you consult ophthalmologist.

Swallowing of acid

1. Ask patient to drink 2 white of egg mixed in water or milk, alternatively ask him to drink soap water.
2. Ask him to gargle with soap water as well.
3. Ask him to drink 500 mL of normal water as well.
4. Apply 2% sodium bicarbonate to lips and tongue, if they are also burnt.

Exposure to Alkali

Commonly used alkalis in biochemistry laboratory are: Sodium hydroxide, ammonium hydroxide, and potassium hydroxide.

Exposure to skin

1. Wash with large quantity of water.
2. Apply 5% acetic acid with cotton wool.

Exposure to eye

1. Spray large quantity of water in eye.
2. Add 3 to 4 drops of saturated boric acid in eye every 5 minutes till you consult ophthalmologist.

Swallowing of alkali

1. Ask patient to drink lemon juice or diluted vinegar (1:3/vinegar: water).
2. Ask him to gargle with same acid solution.
3. Ask him to drink 500 mL of normal water as well.
4. Apply 5% acetic acid solution to lips and tongue, if they are also burnt.

BIOMEDICAL WASTE (BMW) MANAGEMENT

Difference between Hospital Waste and Biomedical Waste

Hospital waste: It refers to all waste, biological or non-biological that is discarded and not intended for further use.

Biomedical waste: It is defined as **"any solid, fluid and liquid or liquid waste, including its container and any intermediate product, which is generated during the diagnosis, treatment or immunization of human being or animals".**

According to WHO:

- Nearly 85% of all waste generated by hospital is general waste.
- About 15% waste is biomedical waste, which includes:
  - Infectious waste—10%.
  - Non-infectious waste such as radioactive and chemical wastes—5%.

Why there is a Need of BMW Management?

Hospitals generate substantial quantity of waste that has potential to cause health and environmental hazards. Safe and sustainable management of biomedical waste (BMW) is social and legal responsibility of all people supporting and financing healthcare activities.

The need for effective biomedical waste management is due to following risks:

1. **Risk of infection** outside hospital for waste handlers, scavengers and at times, general public living in the vicinity of the hospitals.
2. **Injuries from sharps** leading to infection to all categories of hospital personnel and waste handlers.
3. **Risk associated with hazardous chemicals** and drugs to persons handling wastes at all levels.
4. Nosocomial infections in patients from poor infection control practices and poor waste management.
5. **Risk of recycling of "disposables"** which are being repacked and sold.
6. Risk of air, water and soil pollution directly due to waste, or due to defective incineration, emissions and ash.

On March 28, 2016, the Government of India published the **"Biomedical Waste Management Rules, 2016"** in supersession of the Biomedical Waste (Management and Handling) Rules, 1998.

Salient Features of BMW Management Rules, 2016 along with Biomedical Waste Management (Amendment) Rules, 2018

1. The scope of the rules has been expanded to include vaccination camps, blood donation camps, surgical camps or any other healthcare activity.
2. Phase-out the use of chlorinated plastic bags, gloves and blood bags within two years of notification of BMW Management 2016 Rules, i.e. by 27th March, 2018.
3. But as per the Bio-Medical Waste Management (Amendment) Rules, 2018, use of chlorinated plastic bags (excluding blood bags) and gloves has to be phased out by the 27th March, 2019.
4. Pre-treatment of the laboratory waste, microbiological waste, blood samples and blood bags through disinfection sterilization on-site in the manner as prescribed by WHO or NACO.
5. Provide training to all its healthcare workers and immunize all health workers regularly against diseases like tetanus and hepatitis B.
6. Establish a Bar-Code System for bags or containers containing biomedical waste for disposal within one year of notification of rules, i.e. 27th March, 2017. But as per the Bio-Medical Waste Management (Amendment) Rules, 2018, Bar-Code System has to be established in accordance with the guidelines issued by the Central Pollution Control Board by 27th March, 2019.
7. Report major accidents like needle stick injuries, broken mercury thermometer, accidents caused by fire, blasts during handling of biomedical waste and the remedial action taken.
8. Procedure to get authorization is simplified.
9. The new rules prescribe more stringent standards for incinerator to reduce the emission of pollutants in environment.
10. No hospital/healthcare facility (occupier) shall establish on-site treatment and disposal facility, if a service of "common biomedical waste treatment facility" (CBMWTF) is available at 75 km.
11. Operator of a common biomedical waste treatment and disposal facility to ensure the timely collection of biomedical waste from the healthcare facility and assist the healthcare facility in conducting training.

Steps in the management of biomedical waste include:

a. Generation
b. Segregation
c. Collection
d. Storage
e. Treatment
f. Transport
g. Disposal.

Colour Coding of Biomedical Wastes and their Disposal

Biomedical Waste Management Rules, 2016 has categorized the biomedical waste generated from the healthcare facility into four categories based on the segregation pathway and colour code.

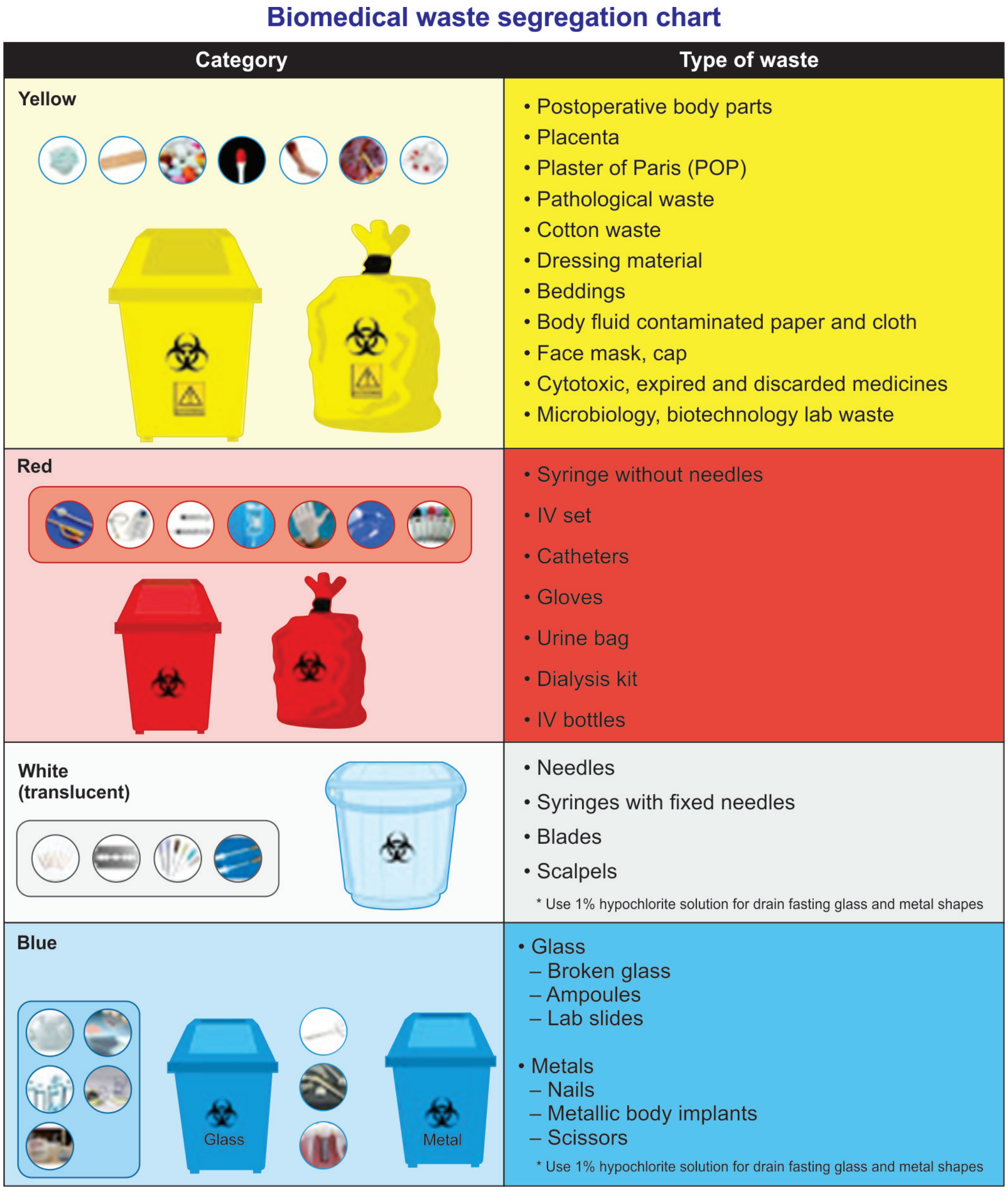


Fig. 2.1: Color coding of biowaste container

Various types of biomedical waste are further assigned to each one of the categories, as detailed below (Fig. 2.1):

1. Yellow category
2. Red category
3. White category
4. Black category

Biomedical wastes' categories and their segregation, collection, treatment, processing and disposal options are summarized in Table 2.1.

Table 2.1: Biomedical wastes' categories and their processing

Category	*Type of waste*	*Type of bag or container to be used*	*Treatment and disposal options*
Yellow	a. **Human anatomical waste**	Yellow-coloured non-chlorinated plastic bags	Incineration or plasma pyrolysis or deep burial
	b. **Animal anatomical waste**		
	c. **Chemical liquid waste:** Liquid waste generated due to use of chemicals in production of biological and used or discarded disinfectants, silver X-ray film developing liquid, discarded formalin, infected secretions, aspirated body fluids.		
	d. **Soiled waste:** Items contaminated with blood, body fluids like dressings, plaster casts, cotton swabs and bags containing residual or discarded blood and blood components.		Incineration or plasma pyrolysis or deep burial* In absence of above facilities, autoclaving or microwaving/ hydroclaving followed by shredding or mutilation or combination of sterilization and shredding.
	e. **Expired or discarded medicines:** Pharmaceutical waste like antibiotics, cytotoxic drugs including all items contaminated with cytotoxic drugs along with glass or plastic ampoules, vials, etc.	Yellow-coloured non-chlorinated plastic bags or containers	Expired cytotoxic drugs and items contaminated with cytotoxic drugs to be returned back to the manufacturer or supplier for incineration at temperature >1200°C
	f. Discarded linen, mattresses, beddings contaminated with blood or body fluid	Non-chlorinated yellow plastic bags or suitable packing material	Non-chlorinated chemical disinfection followed by incineration or plazma pyrolysis or for energy recovery. In absence of above facilities, shredding or mutilation or combination of sterilization and shredding.
	g. **Microbiology, biotechnology and other clinical laboratory waste:** Blood bags, laboratory cultures, stocks or specimens of microorganisms, live or attenuated vaccines, human and animal cell cultures used in research, industrial laboratories	Autoclave safe plastic bags or containers	Pre-treat to sterilize with non-chlorinated chemicals on-site as per National AIDS Control Organization or World Health Organization guidelines thereafter for incineration

(Contd.)

Table 2.1: Biomedical wastes' categories and their processing *(Contd.)*

Category	*Type of waste*	*Type of bag or container to be used*	*Treatment and disposal options*
Red	**Contaminated waste (recyclable):** Wastes such as tubing, bottles, intravenous tubes and sets, catheters, urine bags, syringes (without needles and *fixed needle syringes*), vacutainers, gloves	Red-coloured non-chlorinated plastic bags or containers	Autoclaving or microwaving/ hydroclaving followed by shredding or mutilation or combination of sterilization and shredding. Treated material can be recycled
White (trans-lucent)	**Waste sharps including metals:** Needles, syringes with fixed needles, needles from needle tip cutter or burner, scalpels, blades, or any other contaminated sharp object that may cause puncture and cuts	Puncture-proof, leak-proof, tamper-proof containers	Autoclaving or dry heat sterilization followed by shredding or mutilation or encapsulation in metal container. Combination of shredding-cum-autoclaving; and sent for final disposal to iron foundries
Blue	a. **Glassware:** Broken or discarded and contaminated glass including medicine vials and ampoules (except those contaminated with cytotoxic wastes) b. **Metallic body implants**	Cardboard boxes with blue-coloured marking Cardboard boxes with blue-coloured marking	Disinfection (by soaking in sodium hypochlorite) or through autoclaving or microwaving or hydroclaving and then sent for recycling

VIVA VOCE

Q1. How will you handle the acid spill on hand of your colleague?

Ans. *Exposure to skin*

1. Wash with large quantity of water.
2. Apply 5% sodium carbonate solution with cotton wool.

Q2. What is to be done in case of accidental ingestion of acid?

Ans. *Swallowing of acid*

1. Ask patient to drink 2 white of egg mixed in water or milk, alternatively ask him to drink soap water.
2. Ask him to gargle with soap water as well.
3. Ask him to drink 500 ml of normal water as well.
4. Apply 2% sodium bicarbonate to lips and tongue, if they are also burnt.

Q3. How will you handle the alkali spill on hand of your colleague?

Ans. *Exposure to skin*

1. Wash with large quantity of water.
2. Apply 5% acetic acid with cotton wool.

Q4. What is to be done in case of accidental ingestion of alkali?

Ans. *Swallowing of alkali*

1. Ask patient to drink lemon juice or diluted vinegar (1:3/vinegar:water).
2. Ask him to gargle with same acid solution.
3. Ask him to drink 500 ml of normal water as well.
4. Apply 5% acetic acid solution to lips and tongue, if they are also burnt.

Q5. What is biomedical waste?

Ans. *Biomedical waste* is defined as "any solid, fluid and liquid or liquid waste, including its container and any intermediate product, which is generated during the diagnosis, treatment or immunization of human being or animals".

Q6. What is the need of effective biomedical waste management?

Ans. The need for effective biomedical waste management is due to the following risks:

1. Risk of infection outside hospital for waste handlers and scavengers and at times, general public living in the vicinity of the hospitals.
2. Injuries from sharps leading to infection to all categories of hospital personnel and waste handlers.
3. Risk associated with hazardous chemicals and drugs to persons handling wastes at all levels.
4. Nosocomial infections in patients from poor infection control practices and poor waste management.
5. Risk of recycling of "disposables" which are being repacked and sold.
6. Risk of air, water and soil pollution directly due to waste, or due to defective incineration, emissions and ash.

Q7. How to discard broken glass ampoules?

Ans. Broken or discarded and contaminated glass including medicine vials and ampoules (except those contaminated with cytotoxic wastes.)

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