INTRODUCTION

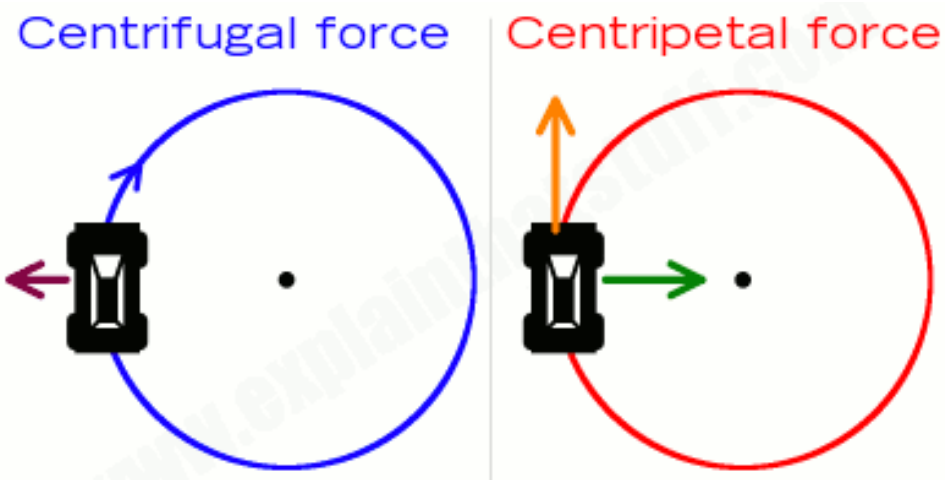
- The driving forces for settling and filtration are gravity and pressure gradients. These forces are usually not enough to create rapid separation.

  Rate = Driving Force / Resistance

- This relationship shows that in order to increase the rate of separation via settling and filtration, is to either:
  1. Decrease resistance
  2. Increase driving force

Centrifugal force

- An object whirled about an axis or center point at a constant radial distance from the point is acted on by a force.
- The object is constantly changing direction and is thus accelerating, even though the rotational speed is constant
- This centripetal force acts in a direction toward the center of rotation
- In cylindrical container, the contents of fluid and solids exert an equal and opposite force, called *centrifugal force*, outward to the walls of the container



The **centrifugal force** on a particle that is constrained to rotate in a circular path is given by

$$F_c = mr\omega^2$$

F_c -centrifugal force.
r - radius of the path,
m - mass of the particle,
W (omega) - **angular velocity of the particle**.

since $\omega = v/r$, where v is the **tangential velocity of the particle**

$$F_c = (mv^2)/r$$

Rotational speeds are normally expressed in revolutions per minute, so

$\omega = 2\pi N/60$ (as it has to be in s^{-1}, divide by 60)

$$F_c = mr(2\pi N/60)^2 = 0.011\, mrN^2$$

where N is the rotational speed in revolutions per minute.

If this is compared with the force of gravity (F_g) on the particle, which F = mg, it can be seen that the centrifugal acceleration, equal to $0.011\, rN^2$, has replaced the gravitational acceleration, equal to g.

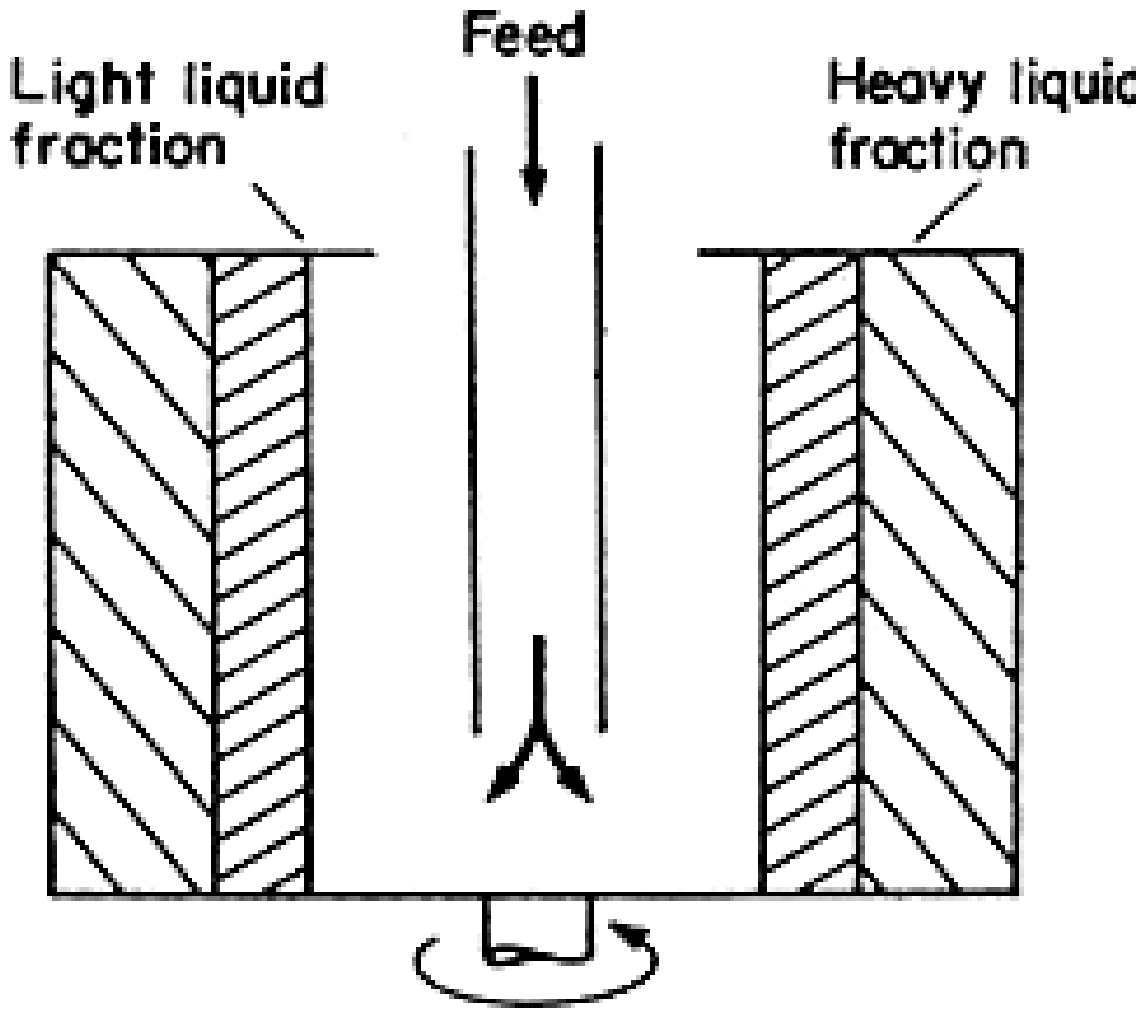
The centrifugal force depends upon

- The radius
- Speed of rotation
- The mass of the particle

In the separation of immiscible liquids (emulsions), the denser liquid moves to the bowl wall and the lighter liquid is displaced to an inner annulus.

The thickness of the layers is determined by

- the density of the liquids,
- the pressure difference across the layers
- the speed of rotation.



A boundary region between the liquids at a given centrifuge speed forms at a radius r_n *where the hydrostatic pressure of* the two layers is equal. This is termed the neutral zone and is important in equipment design to determine the position of feed and discharge pipes. It is found using:

$$r_n^2 = \frac{\rho_A r_A^2 - \rho_B r_B^2}{\rho_A - \rho_B}$$

The subscripts A and B refer to the dense and light liquid layers respectively.

- When particles are removed from liquids in centrifugal clarification, the particles move to the bowl wall under centrifugal force.
- If liquid flow is streamlined the rate of movement is determined by the densities of the particles and liquid, the viscosity of the liquid and the speed of rotation

$$Q = \frac{D^2\omega^2(\rho_s - \rho)V}{18\mu\ln(r_2/r_1)}$$

where $\omega\,(=2\pi N/60)$ = angular velocity, Q ($m^3\ s^{-1}$) = volumetric flowrate, V (m^3) = operating volume of the centrifuge, D (m) = diameter of the particle, ρ_s (kg m^{-3}) = density of particles, ρ (kg m^{-3}) = density of liquid, μ (N s m^{-2}) = viscosity of liquid, r_2 (m) = radius of centrifuge bowl, r_1 (m) = radius of liquid, N (rev s^{-1}) = speed of rotation.

Centrifuge

- **Centrifuge is a device used to separate components of a mixture on the basis of their size, density, the viscosity of the medium, and the rotor speed.**
- The centrifuge is commonly used in laboratories for the separation of biological molecules from a crude extract.
- In a centrifuge, the sample is kept in a rotor that is rotated about a fixed point (axis), resulting in strong force perpendicular to the axis.
- There are different types of centrifuge used for the separation of different molecules, but they all work on the principle of sedimentation.

Centrifugation

- **Centrifugation is the technique of separating components where the centrifugal force/ acceleration causes the denser molecules to move towards the periphery while the less dense particles move to the center.**
- The process of centrifugation relies on the perpendicular force created when a sample is rotated about a fixed point.
- The rate of centrifugation is dependent on the size and density of the particles present in the solution.

Relative Centrifugal Force (RCF)

- Relative centrifugal force is the measure of the strength of rotors of different types and sizes.
- This is the force exerted on the contents of the rotor as a result of the rotation.
- RCF is the perpendicular force acting on the sample that is always relative to the gravity of the earth.
- The RCF of the different centrifuge can be used for the comparison of rotors, allowing the selection of the best centrifuge for a particular function.
- The formula to calculate the relative centrifugal force (RCF) can be written as:

$$\textbf{RCF (g Force)} = 1.118 \times 10^{-5} \times r \times (RPM)^2$$

- where **r** is the radius of the rotor (in centimeters), and **RPM** is the speed of the rotor in rotation per minute.

Centrifuge Rotors

- Rotors in centrifuges are the motor devices that house the tubes with the samples.
- Centrifuge rotors are designed to generate rotation speed that can bring the separation of components in a sample.
- There are three main types of rotors used in a centrifuge, which are:
  - **Fixed angle rotors**
  - **Swinging bucket rotors/ Horizontal rotors**
  - **Vertical rotors**

Fixed angle rotors

These rotors hold the sample tubes at an angle of 45° in relation to the axis of the rotor.

In this type of rotor, the particles strike the opposite side of the tube where the particles finally slide down and are collected at the bottom.

These are faster than other types of rotors as the pathlength of the tubes increases.

However, as the direction of the force is different from the position of the tube, some particles might remain at the sides of the tubes.




Swinging bucket rotors/ Horizontal rotors

Swinging bucket rotors hold the tubes at an angle of 90° as the rotor swings as the process is started.

In this rotor, the tubes are suspended in the racks that allow the tubes to be moved enough to acquire the horizontal position.

In this type of rotors, the particles are present along the direction or the path of the force that allows the particles to be moved away from the rotor towards the bottom of the tubes.

Because the tubes remain horizontal, the supernatant remains as a flat surface allowing the deposited particles to be separated from the supernatant.

Vertical rotors

- Vertical rotors provide the shortest pathlength, fastest run time, and the highest resolution of all the rotors.
- In vertical rotors, the tubes are vertical during the operation of the centrifuge.
- The yield of the rotor is not as ideal, as the position of the tube doesn't align with the direction of the centrifugal force.
- As a result, instead of settling down, particles tend to spread towards the outer wall of the tubes.
- These are commonly used in isopycnic and density gradient centrifugation.




Equipments :

Centrifuges are classified into three groups:

1. Separation of immiscible liquids

2. Clarification of liquids by removal of small amounts of solids (centrifugal clarifiers)

3. Removal of solids (desludging or dewatering centrifuges)

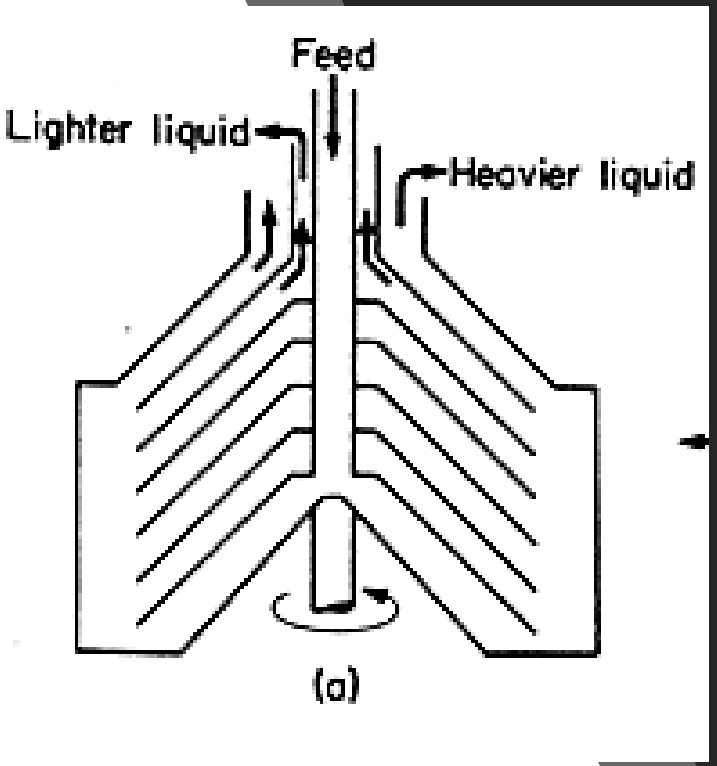
Liquid–liquid centrifuges

- Simplest type of equipment is the ***tubular bowl centrifuge.***

- *It consists of a vertical* cylinder (or bowl), typically 0.1 m in diameter and 0.75 m long, which rotates inside a stationary casing at between 15,000 rpm and 50,000 rpm depending on the diameter.

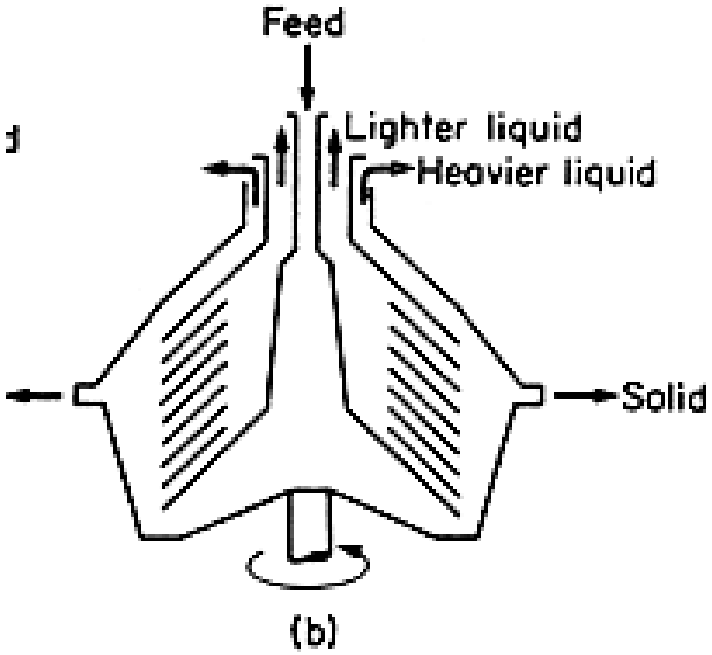
- Feed liquor is introduced continuously at the base of the bowl and the two liquids are separated and discharged through a circular weir system into stationary outlets.

Disk bowl centrifuge



- The feed enters the actual compartment at the bottom and travels upward through vertically spaced feed holes, filling the spaces between the disks
- The holes divide the vertical assembly into an inner section, where mostly light liquid is present, and an outer section, where mainly heavy liquid is present. The heavy liquid flows beneath the underside of a disk to the periphery of the bowl
- The light liquid flows over the upper side of the disks and toward the inner outlet
- Any small amount of heavy solids is thrown outer wall
- Periodic cleaning is required to remove solids deposited
- Disk bowl centrifuges are used in starch-gluten separation, concentration of rubber latex, and cream separation

Centrifugal clarifiers



The **simplest solid–liquid centrifuge** is a *solid bowl clarifier, which has a rotating* cylindrical bowl, 0.6–1.0 m in diameter.

Liquor, with a maximum of 3% w/w solids, is fed into the bowl and the solids form a cake on the bowl wall.

When this has reached a pre-determined thickness, the bowl is drained, and the cake is removed automatically through an opening in the base.

For continuous discharge and feeds which contain a higher solids content are separated using ***nozzle centrifuges or valve discharge centrifuges***.

These are similar to disc bowl types, but the bowls have a biconical shape. In the nozzle type, solids are continuously discharged through small holes at the periphery of the bowl and are collected in a containing vessel.

In the **valve discharge centrifuge** the holes are fitted with valves that periodically open for a fraction of a second to discharge the accumulated solids.

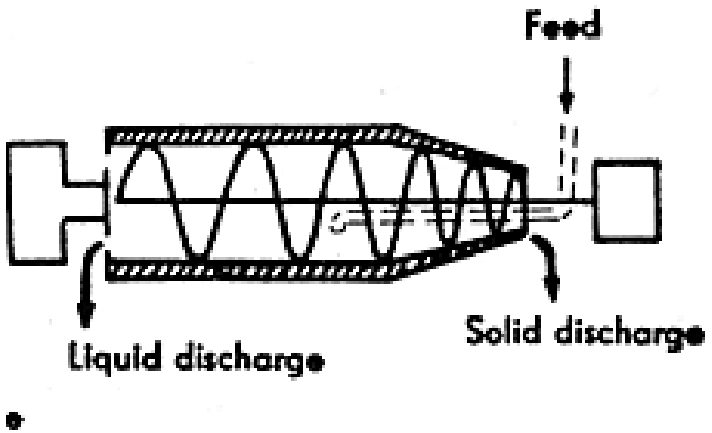
The advantages of this design include less wastage of liquor and the production of drier solids.

Both types are able to separate feed liquor into three streams: a light phase, a dense phase and solids.

Centrifugal clarifiers are used to treat oils, juices, beer and starches and to recover yeast cells.

They have capacities up to 300 000 l h1.

removal of solids

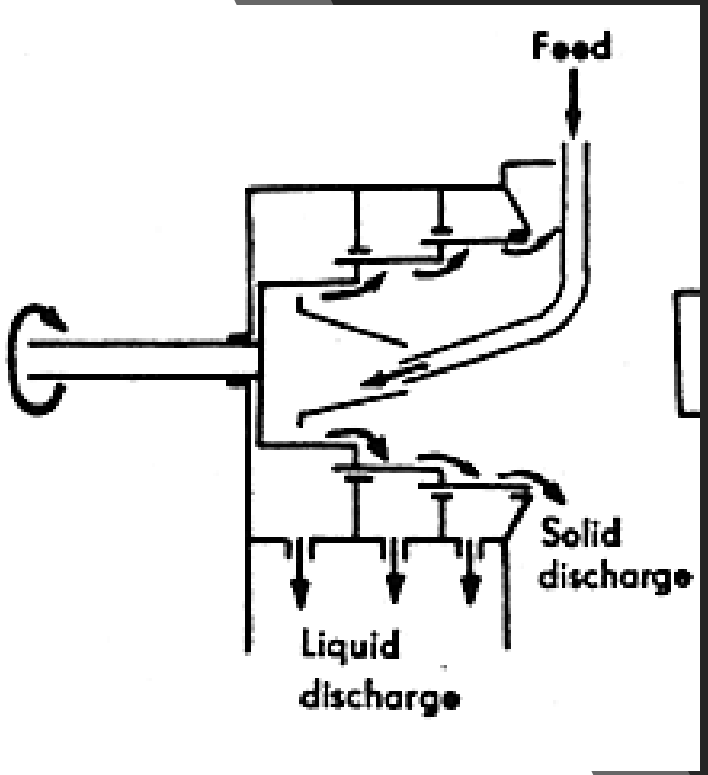


- Feeds with high solids contents are separated using desludging centrifuges, including conveyor bowl, screen conveyor, basket and reciprocating conveyor centrifuges.

- In the ***conveyor bowl centrifuge,*** *the solid bowl rotates up to 25 rpm* faster than the screw conveyor.

- This causes the solids to be conveyed to one end of the centrifuge, whereas the liquid fraction moves to the other larger diameter end.

- The solids are relatively dry compared with other types of equipment.



- The ***screen conveyor centrifuge*** *has a similar design, but the bowl is perforated to* remove the liquid fraction.

- The ***reciprocating conveyor centrifuge*** *is used to separate* fragile solids (for example crystals from liquor). Feed enters a rotating basket, 0.3–1.2 m in diameter, through a funnel which rotates at the same speed.

- This gradually accelerates the liquid to the bowl speed and thus minimizes shearing forces. Liquid passes through perforations in the bowl wall.

- When the layer of cake has built up to 5–7.5 cm, it is pushed forwards a few centimetres by a reciprocating arm. This exposes a fresh area of basket to the feed liquor.

- **Telescoping** method of handling solids from continuous feed is to employ telescoping action in the bowl, sections of the bowl moving over one another and conveying the solids that have accumulated towards the outlet

Benchtop centrifuge

Benchtop centrifuge is a compact centrifuge that is commonly used in clinical and research laboratories.

It is driven by an electric motor where the tubes are rotated about a fixed axis, resulting in force perpendicular to the tubes.

Because these are very compact, they are useful in smaller laboratories with smaller spaces.

Different variations of benchtop centrifuges are available in the market for various purposes.

A benchtop centrifuge has a rotor with racks for the sample tubes and a lid that closes the working unit of the centrifuge.

Continuous flow centrifuge

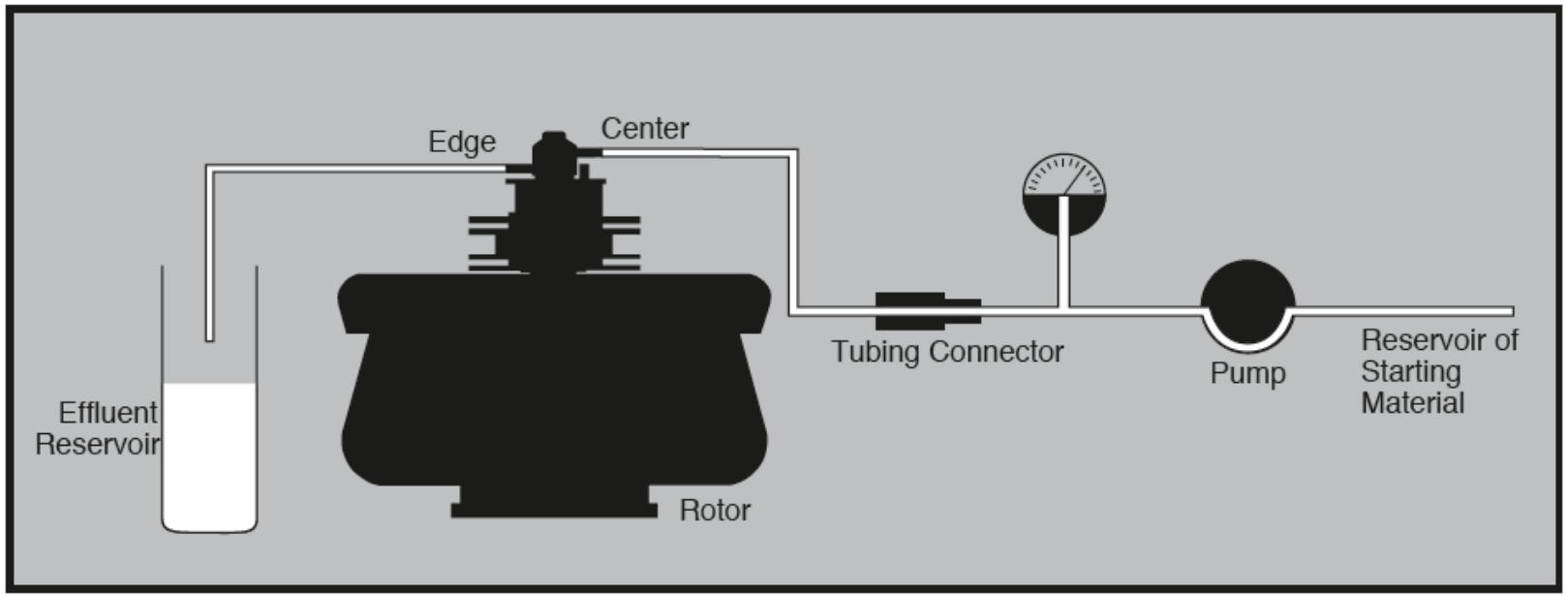
It is a rapid centrifuge that allows the centrifugation of large volumes of samples without affecting the sedimentation rates.

It allows the separation of a large volume of samples at high centrifugal force, thus removing the tedious part of emptying and filling the tubes with each cycle.

They have a shorter pathlength which facilitates the process of pelleting out the solid part out of the supernatant, thus maintaining the speed of the process.

They also have larger capacities which saves time as the sample doesn't have to be load and unloaded over and over again like in traditional centrifuges.

Up to 1 liter of samples can be centrifuged by this centrifuge at a time period of 4 hours or less.



High-speed centrifuge

- The speed of the high-speed centrifuge can range from 15,000 to 30,000 rpm.
- The high-speed centrifuge is commonly used in more sophisticated laboratories with the biochemical application and requires a high speed of operations.
- High-speed centrifuges are provided with a system for controlling the speed and temperature of the process, which is necessary for the analysis of sensitive biological molecules.
- The high-speed centrifuges come with different adapters to accommodate the sample tubes of various sizes and volumes.
- All three types of rotors can be used for the centrifugation process in these centrifuges.

Low-speed centrifuge

Low-speed centrifuges are the traditional centrifuges that are commonly used in laboratories for the routine separation of particles.

These centrifuges operate at the maximum speed of 4000-5000 rpm.

These are usually operated under room temperature as they are not provided with a system for controlling the speed or temperature of the operation.

Swinging bucket and fixed angle type of rotors can be used in these centrifuges.

These are easy and compact centrifuges that are ideal for the analysis of blood samples and other biological samples.

The low-speed centrifuge works on the same principle as all other centrifuges, but the application is limited to the separation of simpler solutions.

Microcentrifuge

- Microcentrifuges are the centrifuges used for the separation of samples with smaller volumes ranging from 0.5 to 2 µl.
- Microcentrifuges are usually operated at a speed of about 12,000-13,000 rpm.
- This is used for the molecular separation of cell organelles like nuclei and DNA and phenol extraction.
- Microcentrifuges, also termed, microfuge, use sample tubes that are smaller in size when compared to the standard test tubes used in larger centrifuges.
- Microcentrifuges with temperature controls are available for the operation of temperature-sensitive samples.

Refrigerated centrifuges

- Refrigerated centrifuges are the centrifuges that are provided with temperature control ranging from -20°C to -30°C.
- Refrigerated centrifuges have a temperature control unit in addition to the rotors and racks for the sample tubes.
- These centrifuges provide the RCF of up to 60,000 xg that is ideal for the separation of various biological molecules.
- These are typically used for collecting substances that separate rapidly like yeast cells, chloroplasts, and erythrocytes.
- The chamber of refrigerated centrifuge is sealed off from the outside to meet the conditions of the operations.

Ultracentrifuges

- Ultracentrifuges operate at extremely high speeds that allow the separation of much smaller molecules like ribosomes, proteins, and viruses.
- It is the most sophisticated type of centrifuge that allows the separation of molecules that cannot be separated with other centrifuges.
- Refrigeration systems are present in such centrifuges that help to balance the heat produced due to the intense spinning.
- The speed of these centrifuges can reach as high as 1,50,000 rpm.
- It can be used for both preparative and analytical works.
- Ultracentrifuges can separate molecules in large batches and in a continuous flow system.
- In addition to separation, ultracentrifuges can also be used for the determination of properties of macromolecules like the size, shape, and density.

Vacuum centrifuge/ Concentrators

- Vacuum centrifuge utilizes the centrifugal force, vacuum and heat to speed up the laboratory evaporation of samples.
- These centrifuges are capable of processing a large number of samples (up to 148 samples at a time).
- This type of centrifuge is used in chemical and biological laboratories for the effective evaporation of solvents present in samples, thus concentrating the samples.
- These are commonly used in high throughput laboratories for samples that might have a large number of solvents.
- A rotary evaporator is used to remove the unnecessary solvents and eliminate solvent bumping.
- The centrifuge works by lowering the pressure of the chamber, which also decreases the boiling point of the samples.
- This causes the solvents to be evaporated, concentrating the particles to be separated.

Further learning…

- https://www.youtube.com/watch?v=XdoCXQ2GOTc
- https://www.youtube.com/watch?v=BaHeehQwVVM
- https://www.youtube.com/watch?v=MRfQYdneGho

Thank you