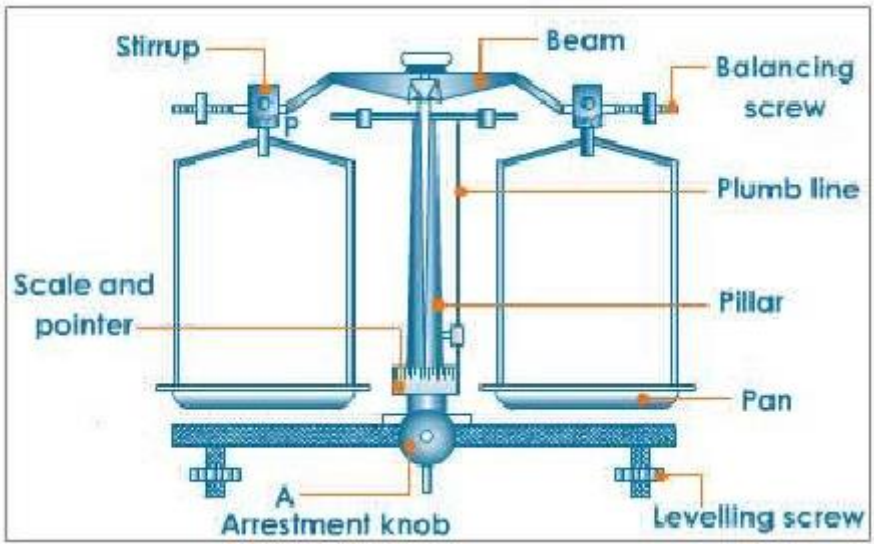
Unit -I. Review of physics

Balance physical & chemical balance. Sensitivity of balance use and care of the balance, mass –volume- specific gravity- units and measurements- properties of matter – viscosity of both fluids- diffusion and osmosis –dynamics- motion – types centripetal force and centrifugal force. application centrifuge principle and parts applications in medicine preventive maintenance ph meter parts and principle cell counter – basic principle.

1.1. PHYSICAL BALANCE



Construction

It is an instrument used to measure the mass of an unknown object using known mass. Unit of mass is Kg (a scalar quantity).It consists of a central beam made of brass or aluminium. The ends of the beam are fixed with balancing screws for adjustment. Two knife edges are fixed in upward position on which rest stirrups which acts as scale pan suspension support. In the middle of the beam there is a central knife edge which points vertically downward. There is a central pillar known to fix at the bottom layer. At the bottom of the wooden base there are levelling screws. A plumb line which is used to set the balance in the horizontal plane is suspended from beam support. A pointer is fixed in vertically downward direction. The pointer moves over the graduated scale attached to the central pillar. Two scale pans of equal masses are suspended from the stirrups. This setup is cased with a glass doors.

Working

Using the physical balance adjust the levelling screws such that the plumb line points vertically downward. Gently raise the central rod by turning the lever. If the pointer remains at the zero mark or oscillates equally on either side of zero mark then the balance is fully adjusted however if the pointer swings more in one direction then adjust the balancing screws such that it swings equally on either side of the zero mark. Lower the object to be weighed in the LHS and place the standard weights in RHS pan till the oscillation reaches about the zero mark. When the central rod is raised use forceps provided in the weight box. Record the mass placed in RHS pan.

Physical balance is based on the principle of moments according to which, when a body is in equilibrium, under the action of a number of forces acting on it in the same plane, the sum of the clockwise moments is equal to the sum of the anticlockwise moments.

For a physical balance with both arms of equal lengths,

Weight on left pan = Weight of standard masses in the right pan.

Since g is constant at the place

Mass of the object in the left pan = mass of standard weights in the right pan.

1.2 SENSITIVITY AND CARING OF BALANCE

1. The weights should be carried with forceps to avoid the change in weight due to moisture and dust particles.
2. The weights and the body whose mass is to be found should be dry.
3. When the pointer is near zero mark, try the weights in the descending order.
4. The beam should be lowered each time before adding or removing weights from the pan.
5. The lever should be turned gently in order to prevent the knife edges from breaking.

Hint : https://www.youtube.com/watch?v=b5K8jHLqOxg

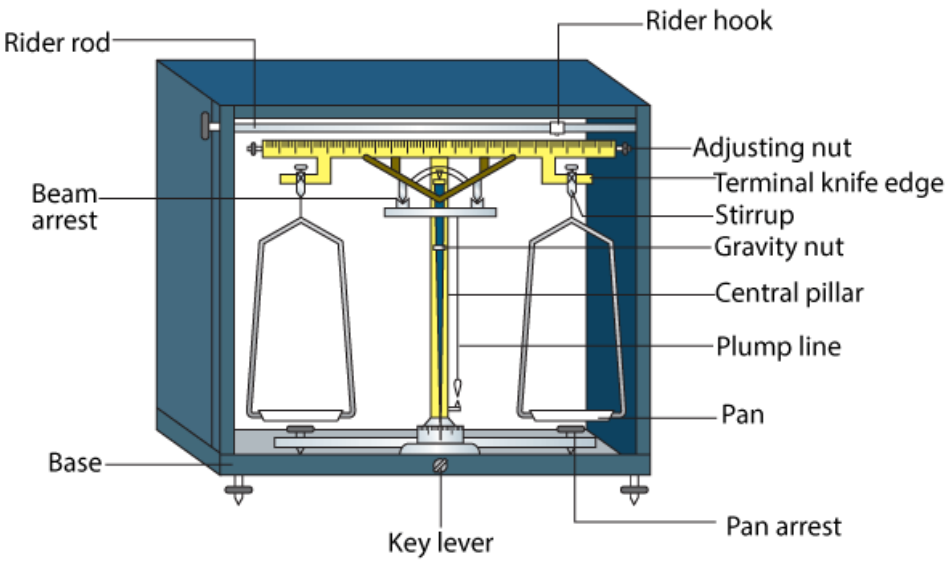
1.3 CHEMICAL BALANCE

The laboratory instrument built for measuring weight is called the balance. The name is derived from mechanical devices that utilize known weights to balance the object to be weighed across a fulcrum. Quantitative analysis involves weighing with great accuracy. Analytical balance in quantitative analysis can be used for weighing substances not heavier than 200g. The least count is 0.0002g.

An electronic balance is one that uses an electromagnet to balance the object to be weighed on a single pan. A chemical can be conveniently weighed on a piece of weighing paper without having to determine the weight of the paper. Taring means that the balance is simply zeroed with the weighing paper on the pan.

The balance is enclosed in a glass case which protects it from dust, air movements etc. The base of the balance rests on screws whereby the edges and agate plates on which they rest are brought into horizontal position by means of plumb bob attached to the balance column. The balance pans are made of light metal coated with nickel to prevent oxidation. Substances should not be placed on pans directly. It should be weighed on watch glasses or in weighing bottles.

A balance that is used to obtain four or five digits to the right of the decimal point in the analytical laboratory is called the analytical balance. The modern laboratory utilizes single pan electronic analytical balance almost exclusively for precision. In the weight box weights are arranged in the order of 50, 20, 20, 10, 5, 2, 2, 1 and it also has a rider made of thin wire. The weight of rider is 0.0002g. The rider is placed on the beam with the help of a hook which is fitted with the balance.



Caring of the Balance

1. Never near the thumb of the pipette. Close the index finger to it.
2. Do not knock out the solution's last drop from the pipette's jet end.
3. Always hold the pipette's lower end in the liquid while being sucked.
4. Fill the burette with the solution carefully and see that the stopcock is not leaking.
5. Carefully handle appliances and chemicals.
6. To read, position the eye precisely on the solution's meniscus level.

1.4 MASS, VOLUME AND SPECIFIC GRAVITY

The **mass** of a body is the quantity of matter contained in it and is an inherent, invariant and independent of neighbouring bodies or place where the body is happens to be situated. Whereas **weight** of the body is the force with which it is attracted by the earth towards its centre and equal to the product of the mass and the acceleration due to gravity.

The **volume** of a body is the space occupied by it. The volume of regular shaped solid bodies can be determined easily by applying appropriate formula. For determining the volume of an irregular solid, a measuring cylinder is used. The measuring cylinder is also used to measure the volume of a liquid.

A measuring cylinder is a cylindrical vessel with graduations in millilitre or cubic centimetre. In order to measure the volume of a given liquid, it is poured in an empty cylinder and the marking corresponding to the upper surface of the liquid is noted, which gives the volume of the liquid. The volume of an irregular shaped solid can be determined by immersing it into water, contained in a measuring cylinder. For example to measure the volume of a stone, the stone is tied with the thread and is lowered into the measuring cylinder. The water level rises. The volume of the stone equals the difference between the volume of water before and after the stone is immersed.

The **specific gravity** is the ratio between the density of an object, and a reference substance. The specific gravity can tell us, based on its value, if the object will sink or float in our reference

substance. Usually our reference substance is water which always has a density of 1 gram per millilitre or 1 gram per cubic centimetre.

1.5 UNITS AND MEASUREMENTS

In mechanics, quantities like length, mass, volume and pressure are called physical quantities. The units which is derived from the fundamental physical quantities such as length (m), mass (kg) and time (sec) are called **fundamental units**.

Different scales of fundamental units

Prefix	Symbol	Factor
tera	T	10^{12}
giga	G	10^{9}
mega	M	10^{6}
kilo	k	10^{3}
deci	d	10^{-1}
centi	c	10^{-2}
milli	m	10^{-3}
micro	μ	10^{-6}
nano	n	10^{-9}
pico	p	10^{-12}

The units which are derived from these physical quantities are called **derived units.**

Eg: Area (m^2), Velocity (m/s), Density (kg/m^3), etc.

There are three principal system of units formulated by International Bureau of Weights and Measures:

(i) **The Centimetre-Gramme-Second (CGS) system:**

Here unit of length is centimetre, unit of mass is gramme and time is second

(ii) **The Foot-Pound-Second (FPS) system:**

Here unit of length is foot, unit of mass is pound and time is second

(ii) **The Metre-Kilogramme-Second (MKS) system:**

Here unit of length is metre, unit of mass is kilogramme and time is second

The first one is used invariable for scientific work, second one in Britain and the third is adopted in electrical engineering, etc., which is found more convenient and useful.

There are six basic units and 2 supplementary units framed by System International (S.I. Units) at 1991, which is applicable and convenience for practice. They are

Sl.No	Physical quantity	S.I. Unit	Symbol
1	Length	Metre	m
2	Mass	Kilogram	kg
3	Time	Second	sec
4	Current	Ampere	A
5	Temperature	Kelvin	K
6	Luminous intensity	Candela	Cd
Supplementary units			
7	Plane angle	Radian	rd
8	Solid angle	Steradian	sr

Rules and conventions of S. I system

1. Unit which containing scientific name should be represented by its first letter in capitals. Eg: Newton- N
2. The symbol for other units should be represented in small letter (Eg: meter – m).
3. Only singular form of unit is used. Avoid punctuation, full stop etc.
4. In Kelvin scale of temperature, degree symbol should not be used.

Dimensions

The fundamental unit of a physical quantity is represented in dimensions. i.e., Fundamental units of length, mass and time is represented by L,M and T respectively.

Physical quantity	Dimensional formula
1. Area = (length)^2	$M^0L^2T^0$, or simply $[L^2]$
2. Volume = (length)^3	$M^0L^3T^0$, or simply $[L^3]$
3. Velocity = length/time	L/T, or M^0LT^{-1}, or $[LT^{-1}]$
4. Acceleration = velocity/time	$\frac{L}{T\times T}=\frac{L}{T^2}$ or M^0LT^{-2} or $[LT^{-2}]$
5. Momentum = (mass × velocity)	$M\times L/T$, or $[MLT^{-1}]$
6. Force = (mass × acceleration) = rate of change of momentum	$\frac{M\times L}{T\times T}=\frac{M\times L}{T^2}$, or $[MLT^{-2}]$
7. Work* = (force × distance or length)	$\frac{M\times L\times L}{T^2}=[ML^2T^{-2}]$
8. Couple* = (force × length)	$M\times(L/T^2)\times L=[ML^2T^{-2}]$
9. Kinetic Energy* = ($\frac{1}{2}$ mass × velocity^2)	$M\times[L^2/T^2]=[ML^2/T^2]$ or $[ML^2T^{-2}]$
10. Potential Energy* = (mass × acceleration due to gravity × distance)	$M\times\frac{L}{T^2}\times L=[ML^2T^{-2}]$
11. Power (*or rate of doing work*) = work/time	ML^2T^{-2}/T, or $[ML^2T^{-3}]$
12. Density = mass/volume	M/L^3, or $[ML^{-3}T^0]$ or $[ML^{-3}]$
13. Specific gravity = a *mere ratio*.	*No dimensions*
14. Pressure = force/area	MLT^{-2}/L^2, or $[ML^{-1}T^{-2}]$
15. Stress = force/area	MLT^{-2}/L^2, or $[ML^{-1}T^{-2}]$

1.6 PROPERTIES OF MATTER

There are three states of matter in the universe such as solid, liquid and gases. **Solids** have definite shape and size. Here the molecules are closed packed. Due to large cohesive force, the molecules are fixed at its original position having freedom to vibrate about this position. **Liquids** has definite volume but no shape of its own. Due to less cohesive force than solid, they are capable to flow and easily compressed. The average distance between two molecules of a liquid is estimated to be more or less of the same order as the size of its molecule. In **gases**, cohesive forces are almost negligible under ordinary conditions of temperature and pressure, so that the molecules lie far apart from each other, the average distance between two molecules being about a hundred times the size of a molecule. Liquids and gases are considered as fluids.

Common properties

1. Matter occupy space. The space occupied by a body is volume
2. Mater has mass
3. A body cannot change its state of rest or uniform motion on its own (inertia)
4. Matter offers resistance
5. Matter is divisible
6. When force is applied, dependent on the intermolecular spacing matter are compressible (Solid – No, Liquid –compressible, Gases – highly compressible).
7. The porosity affects the properties of matter
8. Force of attraction between molecules of same substance is cohesive force while the molecules of different substance is adhesive force.

1.7 VISCOSITY OF BOTH FLUIDS

Flow is defined as the quantity of a fluid, i.e. a gas or a liquid, passing a point in unit time. $F = \frac{Q}{t}$

where F = mean flow; Q = quantity (mass or volume); t = time.

Laminar Flow

In laminar flow (Fig. 3) a fluid moves in a steady manner and there are no eddies or turbulence. This is the type of flow normally present in smooth tubes at low rates of flow. The flow is greatest in the centre, being about twice the mean flow, as illustrated by the longer arrows in the figure. As the side of the tube is approached the flow becomes slower until it approaches zero at the wall. In order to drive a fluid through a tube, a pressure difference must be present across the ends.

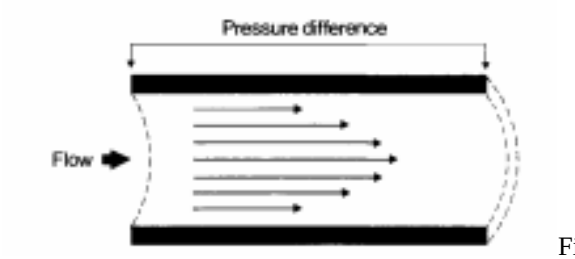


Fig. 3. Laminar flow.

The graph in Fig. 4 shows the result if various flows are passed through a tube and the resulting pressure drop across the ends is recorded. There is a linear relationship so that flow is directly proportional to pressure under conditions of laminar flow. The ratio of pressure to flow is a constant known as the resistance R of the apparatus or tube concerned.

Figure 5 shows how resistance can be measured. A known constant flow Q is passed through the apparatus concerned and the difference in pressure $P_1 - P_2$ between the ends of the apparatus is measured. By dividing the pressure difference by the flow the resistance of the apparatus is obtained and, provided that flow is laminar, the resistance is independent of the flow. A technique such as this may be used to measure resistance with either gas or liquid flow.

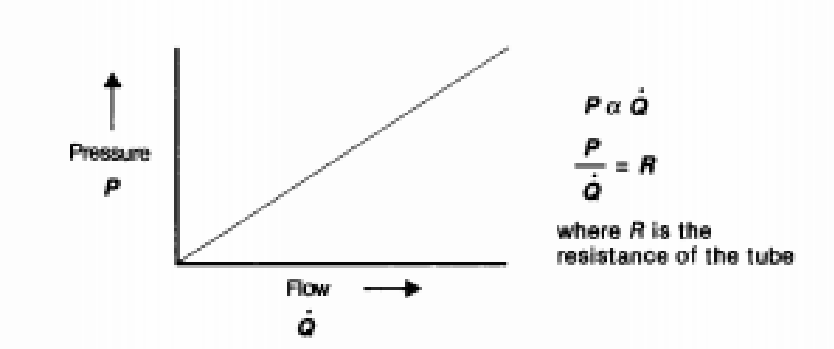


Fig. 4. The graph shows the linear relationship of flow to pressure in laminar flow.

Alternatively, for liquids a constant head of pressure may be achieved by means of a reservoir of liquid and measurement of the resulting flow allows calculations of the resistance. A system of this type may be used to illustrate what happens if the diameter of a tube is halved (Fig. 6).

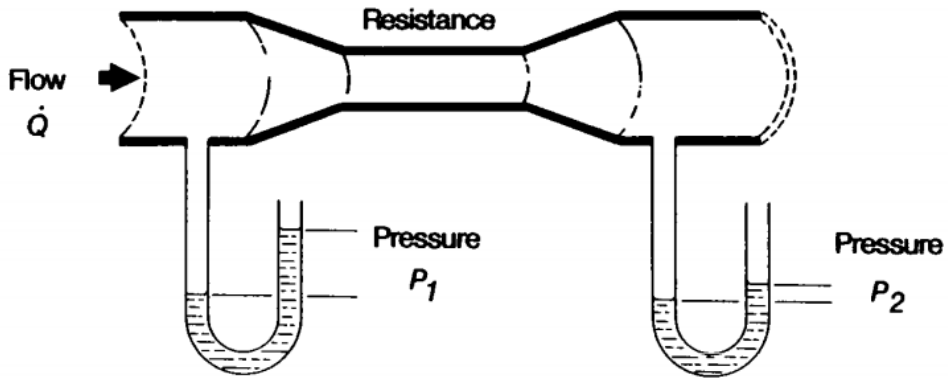


Fig. 5. Measurement of flow-resistance

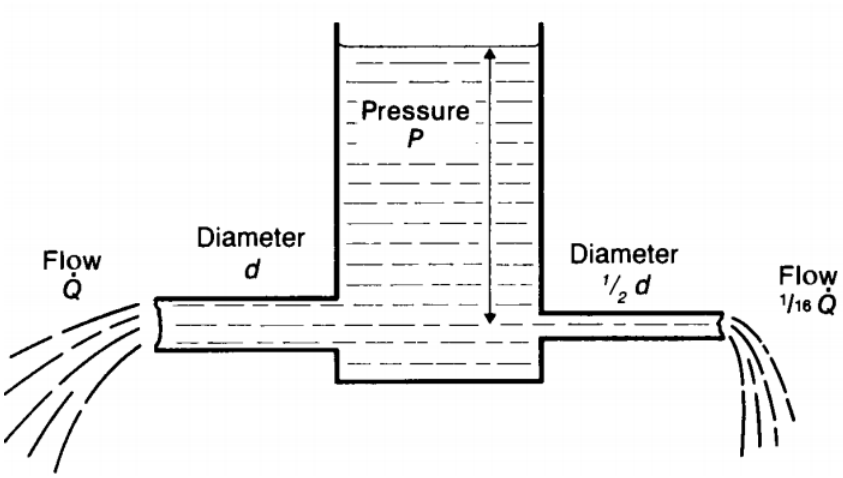


Fig. 6. Effect on flow of halving the tube diameter.

It is found that this reduction of the diameter has a pronounced effect on the resistance to flow. Halving the diameter reduces the flow to one sixteenth of its original value if the pressure drop along the tube remains the same. In other words, the flow is proportional to the fourth power of the diameter. Consequently, a slight reduction of the diameter of an endotracheal tube can have an appreciable effect on resistance and therefore on flow.

As shown in Fig. 7, the effect of altering the length of the tube is much less marked than that of altering the diameter. If the length of the tube is halved, the flow will double, other factors being kept constant. Finally, the viscosity of the fluid affects resistance to laminar flow in such a way that the higher the viscosity the slower is the flow. Viscosity is a measure of the frictional forces acting between the layers of the fluid as it flows along the tube. It is represented by the Greek letter eta (η) and has the units of Pascal seconds. The effects described may be summarized as follows:

$$Q \propto \frac{Pd^4}{l\eta}$$

where Q = flow through tube; P = pressure across tube; d = diameter of tube; l = length of tube; η = viscosity of fluid

All these factors are incorporated in an equation which can be derived theoretically and is known as the Hagen-Poiseuille equation:

$$Q = \frac{\pi P d^4}{128 \eta l}$$

This equation can be rearranged to show how the pressure drop across a tube depends on various factors:

$$P = \frac{128 \eta l Q}{\pi d^4}$$

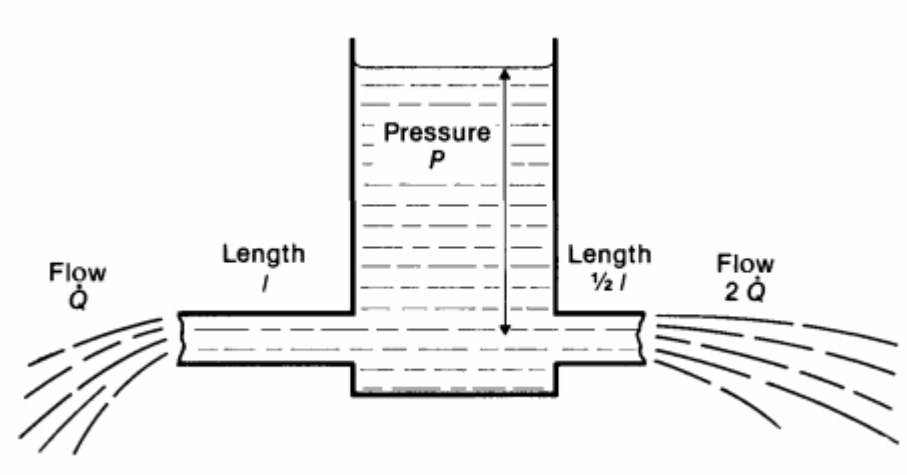


Fig. 7 Effect on flow of halving the length of a tube

Turbulent flow

Laminar flow is not the only type of flow occurring in anaesthetic apparatus, breathing systems, the airways and the circulation. As illustrated in Fig. 8, laminar flow may change to turbulent flow if a constriction is reached which results in the fluid velocity increasing. In turbulent flow, fluid no longer flows in a smooth fashion but swirls in eddies and the resistance is higher than for the same laminar flow. The variation of fluid velocity across the tube is different in turbulent flow from that which occurs in laminar flow and the flow is no longer directly proportional to pressure. The theoretical analysis of turbulent flow is highly complex and although it is possible to predict some aspects of flow behaviour, much of what is known has been learnt from experiments. For turbulent flow in tubes that are rough on the inside it is found that the flow is approximately proportional to the square root of the pressure (Fig. 8), i.e. in order to double the flow, the pressure must be increased by a factor of four. As the relationship of pressure to flow is no longer linear, resistance is not constant and when referring to resistance in the pressure of turbulent flow, it is important that the flow at which the resistance is measured is specified. For example, the resistance to the flow of air during breathing depends on the air flow if the flow is turbulent.

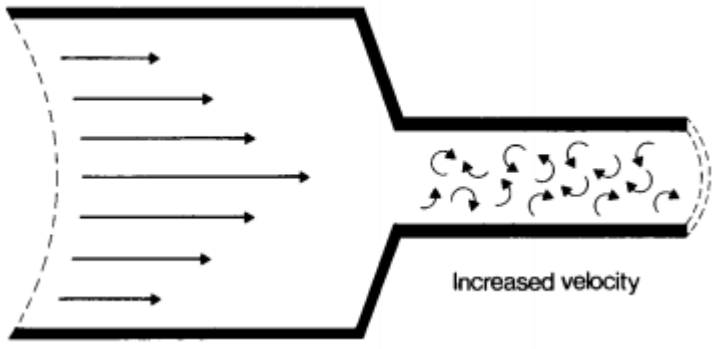


Fig. 7 Onset of turbulent flow

The factors affecting flow and pressure during turbulent flow are as follows:

$$Q \propto \sqrt{\frac{P}{l\rho}} \quad \text{(or)} \quad P \propto Q^2 l \rho$$

Q = flow; P = pressure across; *l* = length of tube; p = density of fluid

The property of the fluid passing through the tube that is important in turbulent flow is the density, which is represented by the Greek letter rho (p) and is equal to mass divided by volume (kgm^{-3}).

The pressure difference necessary to produce a given flow increases as the diameter of the tube decreases, the increase required being slightly greater than that for laminar flow. However it is not expressible as a simple power of the diameter.

For turbulent flow in smooth tubes the resistance shows behaviour intermediate between turbulent flow in rough tubes and laminar flow, so there is some dependence on viscosity as well as density. The behaviour of turbulent flow in smooth tubes is not straightforward so no simple formula can be given to relate pressure drop to the other factors.

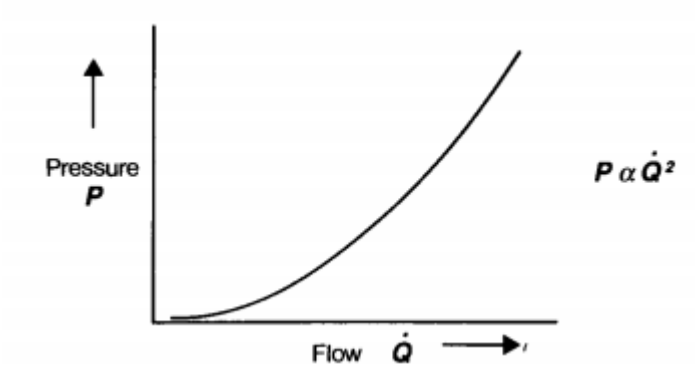


Fig. 8. The graph shows the relationship of flow to pressure in turbulent flow.

1.8 DIFUSSION AND OSMOSIS

Diffusion is the process by which the molecules of a substance transfer through a layer or area such as the surface of a solution. In the lungs, gases diffuse across a gas-liquid barrier in this way, but diffusion may also occur at other sites, shown diagrammatically in Fig. The diffusion of gases in the body is illustrated by compartments in the form of layers in a bottle. At the top, gas in the alveoli of the lungs is represented and below it the alveolar-capillary membrane where the gas diffuses into the liquid phase, i.e. the blood. There are then further membranes at which diffusion occurs, between the blood and the extracellular fluid, and between the extracellular fluid and the cell, allowing the gas to pass from one compartment of the body to the next.

Diffusion can still take place without a membrane or a gas-liquid barrier. For example, if gas escapes from a broken gas pipe, the gas spreads by diffusion even after the gas tap has been turned off.

PRACTICAL ASPECTS OF DIFFUSION

Consider now some practical points regarding diffusion and anaesthetic apparatus. Anaesthetic systems deliver halothane vapour through corrugated rubber tubing and, as halothane is very soluble in rubber it diffuses readily into and through the rubber of the tube. The rubber tube thus provides a possible source from which halothane may diffuse into the theatre atmosphere,

in addition to the more direct route from release of halothane vapour through the expiratory valve. halothane may remain stored in the rubber due to its high solubility, and trace levels may be released later. As another example of the effect of tension gradients on diffusion, consider a patient with decompression sickness in whom bubbles of nitrogen form in the tissues. On the left of Figure is a bubble of nitrogen in the tissues within a capillary loop. If the patient then receives anaesthesia with 50% nitrous oxide, a tension gradient develops as shown and the arterial tension of nitrous oxide could be 50 kPa compared with zero tension within the bubble. Nitrous oxide diffuses into the bubble thus enlarging it, as shown on the right, and aggravates the condition. The problem is not limited to patients with decompression sickness but applies equally to those with bubbles of air at other sites such as a pneumothorax. Even air in the middle ear expands and this may give rise to problems in grafts in that region. In Fig.1 there is a reverse gradient for nitrogen, but the nitrogen is not removed from the bubble at the same rate that nitrous oxide diffuses into it because the lower solubility of nitrogen restricts the rate of its diffusion and the rate at which the capillary bloodstream removes it. Another example of nitrous oxide diffusion arises routinely during the course of anaesthesia, when the gas diffuses into the cuff of an endotracheal tube so increasing the pressure on the trachéal mucosa.

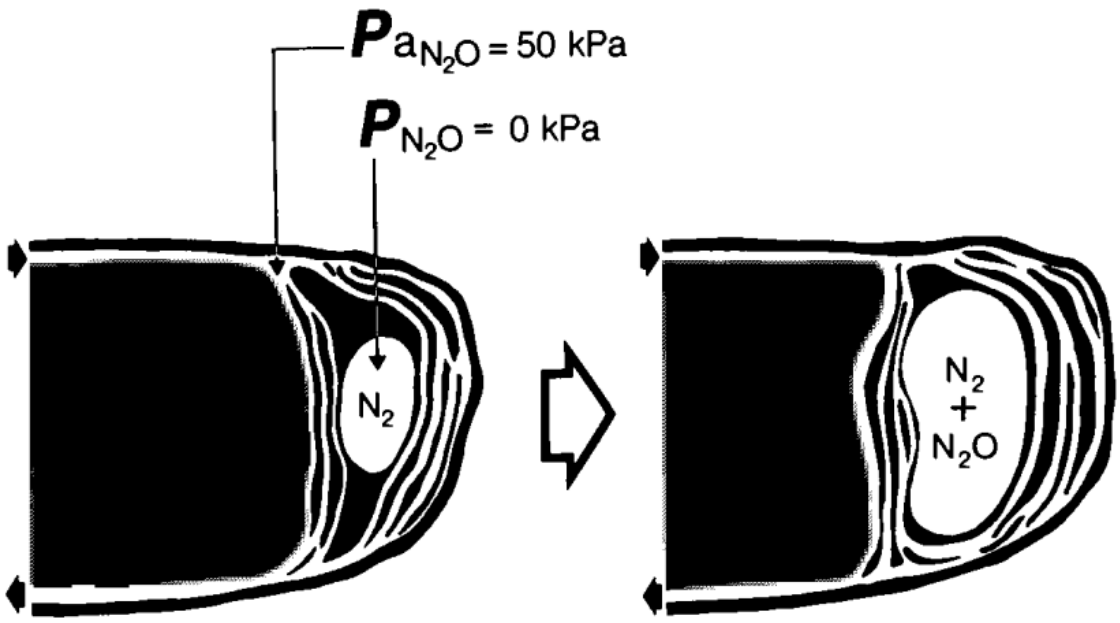


Fig. 1. Effect of nitrous oxide on the size of a bubble of nitrogen in the tissues.

OSMOSIS

Consider now a membrane that does not allow the passage of all molecules, i.e. a semipermeable membrane, separating a solution from a solvent as shown in Fig. 2.

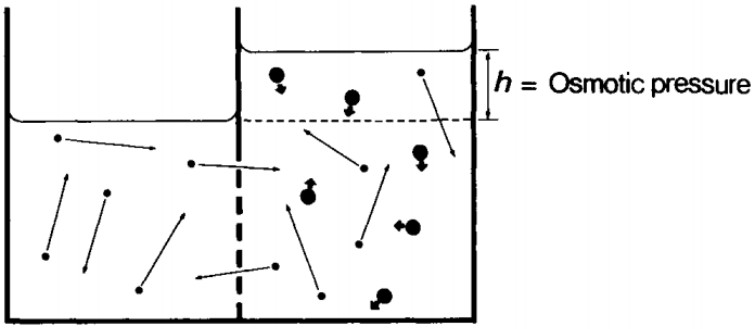


Fig. 2 Principle of Osmosis

The small molecules of solvent can readily pass through the membrane, but in the compartment on therightthere are some large solute molecules which cannot pass through. In this compartment the concentration of small solvent molecules is reduced by the presence of the solute. If Fick's law of diffusion is applied, what would happen to the small molecules? They diffuse from the area of higher concentration on the left across to the right side, giving a transfer of liquid. If the transfer is allowed to continue long enough, equilibrium is achieved and then 'A', the excess height of the compartment on the right, represents the osmotic pressure. It is the pressure which exactly counterbalances the effect of the dissolved molecules. An alternative way of achieving equilibrium is to have a closed compartment as shown in Fig. 3.

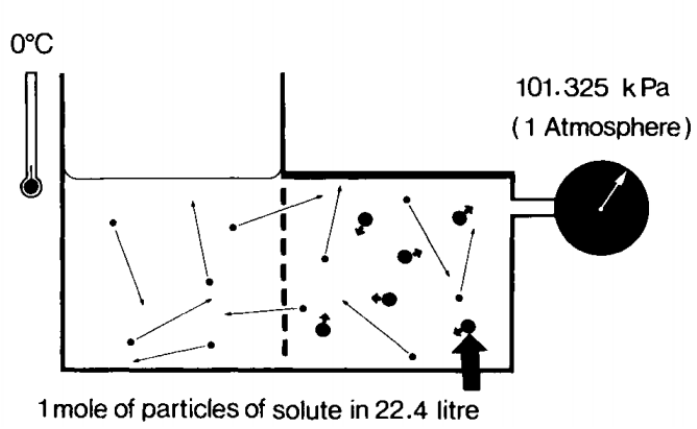


Fig. 3 101.325 kPa osmotic pressure is produced when 1 mol solute is dissolved in 22.4 litres of solution at 0°C.

If the compartment on the right has a 22.4 litre capacity, and if it contains 1 mol of particles of solute at a temperature of 0°C, then the osmotic pressure built up in this compartment is 101.325 kPa or one standard atmosphere. This is shown in the diagram by the gauge pressure, this being additional to the ambient pressure also present. Note that this is similar to the concept that 1 mol of a perfect gas in a volume of 22.4 litres exerts a pressure of one atmosphere. Provided that 1 mol of solute is present in the compartment and that the membrane is not permeable to the solute molecules, the size of these molecules is immaterial.

In clinical practice there are many different types of molecules present in plasma, urine or other body fluids. In order to distinguish between the molarity of individual specific components and the sum total of molarities which give rise to the osmotic pressure the term 'osmolarity' is used. As an example consider the components of Ringer lactate solution.

Ringer lactate solution:

Sodium - 131 mmol litre-1

Potassium - 5 mmol litre-1

Calcium - 2 mmol litre-1

♦Lactate - 29 mmol litre-1 (* Assuming the lactate is all in ionized form.)

Chloride -111 mmol litre-1

Total osmolarity-278 mosmol litre-1

Each component is present in various concentrations of mmol litre-1, but it is the sum total of the molarities which givesriseto the osmotic pressure. In other words, the osmolarity of the solution is the sum of the molarities of the solutes. In Ringer lactate solution it is 278 mosmol litre-1. Most bodyfluidssuch as plasma have an osmolarity of about 300 mosmol litre-1. Over 99% of the osmolarity of plasma is due to electrolytes such as sodium, chloride and bicarbonate, the contribution of the plasma proteins being very small at around 1 mosmol litre-1. Mostfluidsadministered to the patient are adjusted to have an osmolarity of around 300 mosmol litre-1. This means that they are isotonic with body fluids. If a patient is transfused with fluids of low osmolarity, i.e. hypotonie fluids, then the change in the osmotic pressure gradient across the cell membranes causes fluid to diffuse into the cells giving a rise of hydrostatic pressure in them. Use is made of this fact in the red-cell fragility test for the detection of haemolytic anaemias. Red cells are added to various concentrations of saline at 20°C. The lower the osmolarity of the solution, the greater the hydrostatic pressure which builds up in the red cells as fluid passes into them. Thus, at osmolarities below 200 mosmol litre^{-1}, the red cells burst and release haemoglobin into the solution. In many haemolytic anaemias the red cells are abnormal and more fragile and so burst at higher osmolarities than normal. The main difference between plasma and interstitial fluid is the relative proportions of protein, which is negligible in the latter. As the membrane of blood capillaries is permeable to water and electrolytes but not to large protein molecules, the osmotic pressure between the blood and extracellular fluid depends on the protein molecules. To distinguish this pressure from total osmotic pressure it is sometimes called the oncotic pressure. Although albumin and globulin in the plasma have a very small osmolarity of around 1 mosmol litre^{-1} they give rise to an oncotic pressure in the capillaries of about 3.5 kPa (26 mmHg), the pressure being principally due to the albumin because of its higher molar concentration. Figure 5 illustrates a capillary loop with an arterial pressure of 7 kPa (53 mmHg) and a venous pressure of near zero. The balance of pressures is closely related to the oncotic pressure from the plasma proteins. Thus, at the arterial end of the capillary, the hydrostatic pressure exceeds the oncotic pressure by 3.5 kPa and this results in the passage of fluid out of the capillary. At the venous end the pressure ratios are reversed and tissue fluid therefore passes into the capillary. These pressure gradients produce a continual fluid flow through the tissues to help carry oxygen and carbon dioxide and other agents to and from the cells. This is an important mechanism because diffusion alone does not enable solutes to move rapidly through a liquid. The example is simplified as the small hydrostatic and oncotic pressures in the tissue fluid have been disregarded. A fall of plasma oncotic pressure, e.g. to below 2 kPa (15 mmHg), lowers the gradient at the venous end of the capillary and consequently tissue fluid can then accumulate as oedema. Measurement of oncotic pressure may therefore help with the management of

patients with pulmonary or tissue oedema. The instrument used for such measurements is an oncometer and is based on the principle shown in Fig. 4. A semipermeable membrane separates the plasma sample from a saline reference solution, and the change due to oncotic pressure is measured by a transducer.

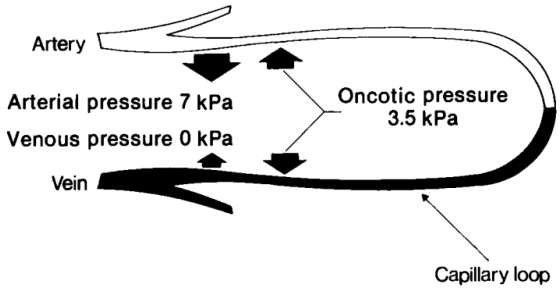


Fig 5.Pressure balance in a capillary loop.

1.9. DYNAMICS

It is a branch of mechanics which deals with objects under motion. It is further classified in to Kinematics and kinetics. Kinematics deals with the motion of objects without bothering about the cause of the motion. Whereas kinetics deals with motion of objects consideration the cause of their motion.

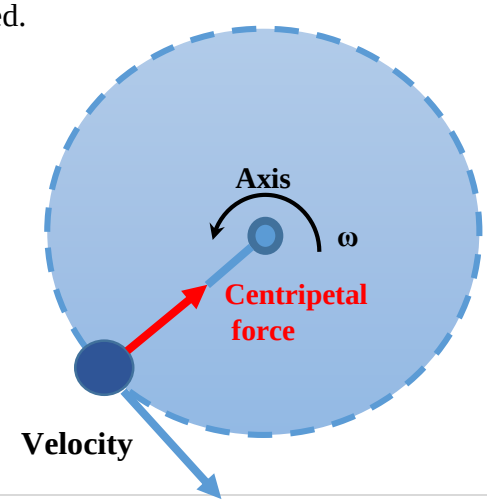
Centripetal force. According to Newton's first law of motion, a body must continue to move with a uniform velocity in a straight line, unless acted upon by a force. straight line, unless acted upon by a force. It follows, therefore, that when a body moves along a circle, some force is acting upon it, which continually deflects it from its straight or linear path ; and, since the body has an acceleration towards the centre, it is obvious that the force must also be acting in the direction of this acceleration, i.e., along the radius, or towards the centre of its circular path. It is called the centripetal force.

Centripetal force = $mv\omega = mv^2/r = mr\omega^2$

Centrifugal force. The equal and opposite reaction to the centripetal force is called the centrifugal force, because it tends to take the body away from the centre. Centripetal and centrifugal forces being just action and reaction in the sense of Newton's third law of motion.

1.10 CENTRIFUGATION – PRINCIPLE, TYPES AND APPLICATIONS

It is a technique of separating substances which involves the application of centrifugal force. The particles are separated from a solution according to their size, shape, density, viscosity of the medium and rotor speed.



1. In a solution, particles whose density is higher than that of the solvent sink (sediment) and particles that are lighter than it float to the top.
2. The greater the difference in density, the faster they move. If there is no difference in density (isopyknic conditions), the particles stay steady.
3. To take advantage of even tiny differences in density to separate various particles in q solution, gravity can be replaced with the much more powerful "centrifugal force" provided by a centrifuge.
4. A centrifuge is a piece of equipment that puts an object in rotation around a fixed axis (spins it in a circle), applying a potentially strong force perpendicular to the axis of spin (outward).
5. The centrifuge works using the sedimentation principle, where the centripetal acceleration causes denser substances and particles to move outward in the radial direction.
6. At the same time, objects that are less dense are displaced and move to the centre.
7. In the laboratory centrifuge that uses sample tubes, while low-density substances rise to the top.

Types of Centrifuges

(1) Low speed centrifuges

(2) High speed centrifuges

(3) Ultracentrifuges

Applications

1. To separate two miscible substances
2. To analyse the hydrodynamic properties of macromolecules
3. Purification of mammalian cells
4. Seperating chalk powder from water
5. Removing fat from milk to produce skimmed milk
6. Separating particles from an air-flow using cyclonic separation
7. The clarification and stabilization of wine, etc.,

MEASURING ACIDITY - PH FUNCTION

Solutions with a pH less than 7 are acidic, and solutions greater than 7 are basic or alkaline. Solutions with a pH of exactly 7 are pH neutral, neither acidic nor basic. You know certain bodily fluids have characteristic pHs. Blood has a pH of 7.4. Stomach juice has a pH of around 1.

The p-Function

The p in pH is a mathematical operator. We have been dealing with several operators, including addition, subtraction, and square roots. The p-function operator isn't that different, but it looks a little strange. The p-function operator means the negative logarithm of. The H in pH means hydrogen ion concentration. So we have a definition of pH:

$pH = - \log [H^+]$

Chemists use the p-function operator to express the concentrations of many ions. pOH for hydroxide ion, pCa for Ca2+, and so forth. The meaning of the p is the same in every case-take the logarithm of the concentration and then change the algebraic sign.

Now, there are very practical reasons for defi ning the p-function. First, the hydrogen ion concentration varies from 10^0 M to 10^{-14} M in solutions commonly encountered in the laboratory. That is 14 orders of magnitude - a factor of a hundred million million. Imagine what a pH meter would look like without the logarithm part of the p-function definition. The scale would have a hundred million million divisions. A logarithm function is a way to map a vast range of values onto a much smaller set of values. In this case, the logarithm function maps a range from 0 to 10–14 onto a range from 1 to 14. The negative sign in the definition is one of convenience. Nobody likes working with negative numbers.

The negative sign simply ensures that the pH values will be positive numbers. To apply the p-function, you simply take the logarithm of the [H+] concentration and then change the sign. For example, what is the pH of a solution?

when the $[H^+]$ is 1.0×10^{-3} M?

$pH = -\log (1.0 \times 10^{-3} \text{ M}) = 3.00$

A couple of comments are in order here. First, did you notice the value of the pH is the same as the absolute value of the exponent? This will always be true when the first part of the scientific notation is exactly 1. The second comment relates to significant figures. There are two significant figures in the molarity measurement of 1.0×10^{-3} M. There are also two significant figures in the pH value of 3.00. Finally, pH values have no intrinsic units. Logarithms represent "pure numbers," and as such, have no units.

Principle cell counting

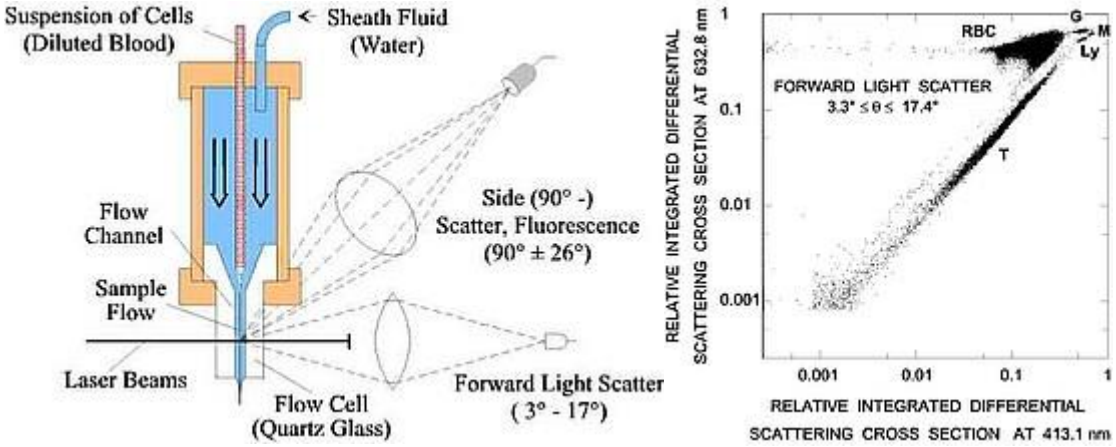
Cells can readily be counted using a microscope. The exact sample volume is usually determined using counting chambers, which have a known height. The sample volume is then derived using the calibrated microscope magnification. However, detailed morphological information is usually not needed for proper identification of cell type. In this case flow cytometry offers advantages over microscopic counting for examination of cell suspensions. A large number of cells can be measured by flow cytometry. This is essential to measure the concentration of rare cells in a mixture (e.g. stem cells circulating in blood). In addition the concentration can be measured with lower statistical uncertainty. Two detection techniques are routinely applied in flow cytometry, laser flow cytometry and impedance counting.

Principle of laser flow cytometry

Blood cells of a diluted measurement suspension are injected into a capillary, and hydrodynamically focussed by a sheath flow to finally enter the flow channel (250 µm x 250 µm) of a quartz cell. The diameter of the sample fluid, which contains the blood cells, is reduced to about 5 µm because of hydrodynamic focussing. Hence the blood cells cross the focus of the laser beam in single file. Intensity of forward light scatter and orthogonal light scatter is measured simultaneously for each blood cell. In addition, laser-induced fluorescence signals are detected when analysing stained blood cells.

In the figure on the right hand side the result from a measurement of a diluted blood sample is shown. The relative intensity of forward light scatter at a wavelength of 632.8 nm is plotted versus forward light scatter at 413.1 nm. In this scatter diagram, each blood cell is represented by a single dot. The clusters indicated in the scatter diagram correspond to red blood cells (RBC), platelets (thrombocytes, T) and subpopulations of leukocytes, i.e. lymphocytes (Ly), monocytes (M) and granulocytes (G).

The axes represent the integrated differential cross section for light scattering, the corresponding angle of observation ranges from 3.3° to 17.4°. For calibration of the cross section monodisperse polystyrene microspheres of known sizes can be used, the integrated cross sections of which are calculated by Mie theory.

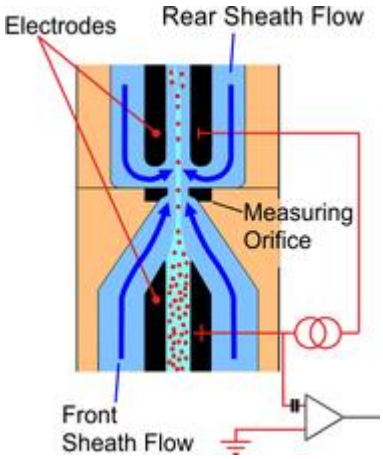


Principle of impedance counting

Blood cells of a diluted blood sample pass a measuring sensor in single file. Typically, the diameter and length of the orifice amounts to 60 µm. The front sheath fluid is used for hydrodynamic focussing of the sample flow, which contains the blood cells. The rear sheath flow is required to avoid recirculation of blood cells after passing the orifice. As sheath fluids, isotonic solution is used. A constant current of up to 1mA is applied between the electrodes. The voltage between both electrodes is changed due to the modified electrical conductivity when a blood cell passes the measuring sensor.

This effect allows detection of single blood cells. The amplitude of the signal is approximately proportional to the particle volume and hence red blood cells (volume about 90 fL) and platelets (volume about 6 fL) can be distinguished. A typical histogram is shown in the figure to the left. Here the number of events is plotted in a histogram, i.e. cell count versus the amplitude of the impedance signal. Two distributions of cells are observed corresponding to red blood cells (erythrocytes) and platelets (thrombocytes).

To enumerate leukocytes by impedance counting erythrocytes are destroyed by lysing reagents. Subsequently the measuring suspension is analysed. A typical resulting pulse height distribution of a control blood sample is shown in the histogram on the right hand side.


Electrodes
Rear Sheath Flow
Measuring
Orifice
Front
Sheath Flow