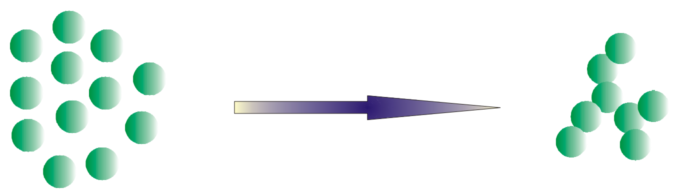
SEDIMENTATION

The nature of aggregate may depend on the conditions of its formation.

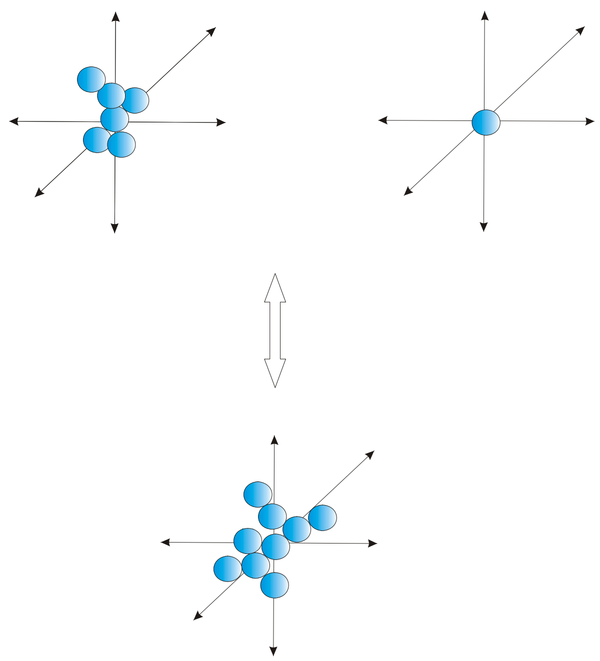
FLOC – open aggregate



COAGULATE – coagulation from denser form of floc
Particles in the floc still maintain individuality ,but still a bad news for colloid scientists.
What initial focculation?
Decrease in total degree of translational freedom → spontaneous

$\therefore\ \Delta S_{(agg)}$ - decreases

$\Delta G_{(agg)} = -T\ (\Delta S_{agg})$ increases



Coming of a particle to an aggregate → loss in the independent translational motion.
ΔG of interaction between particles also deacreases.

Hence, surface tension between interacting surface decreases.

$\therefore$ increases the number of particles in the floc

*will result in **sedimentation.***

SEDIMENTATION

Disperse system :

a) – all particles are the same size

$\Rightarrow$ monodisperse

b) – particles are of a range at sizes.

$\Rightarrow$ polydisperse

What to keep these particles suspended in solution?

Thermal motion

Brownian motion

Under influence of gravity

$\Rightarrow$ particle will separate out

Normally ,

1 μm diameter

relative density ≥ 2.5 - *will separate out*

Newton's Law

$$mg - m'g - fv = \frac{dv}{dt}$$

The fictional force increases with v setting of a particle in a dense medium $\rightarrow$ balances the downwards force,

the acceleration ≈ 0

$\therefore$ travels at v_t (terminal velocity)

assume that spherical

$fv = Bv_t$

$B = 6\pi\eta r$

Stokes settling radius, *r*,

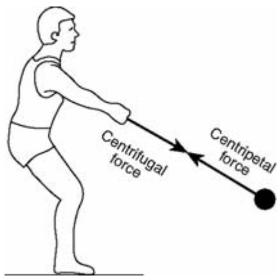
$$(\rho_s - \rho_l)\frac{4}{3}\pi r^3 g = 6\pi\eta v_t r$$

$$v_t = 2\frac{(\rho_s - \rho_1)gr^2}{9\eta}$$

Where, ρ_s – density of the solid particle

ρ_1 – density of the liquid

CENTRIFUGAL FORCE



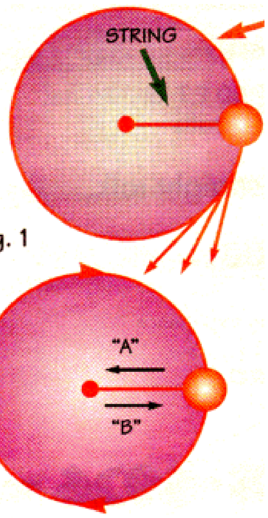
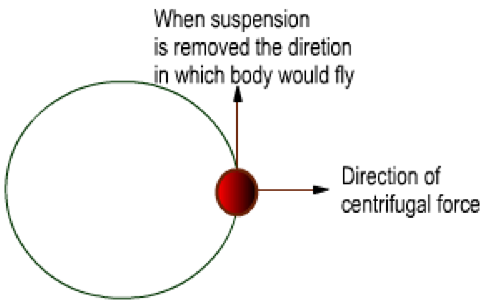


Fig. 1



An object that moves in a circle is undergoing a continual change in direction. This means that objects are constantly being accelerated - always toward the center of rotation.

"A" shows the direction in which CENTRIPETAL force acts.

"B" is CENTRIFUGAL force.

If CENTRIFUGAL force > CENTRIPETAL force, then???

Consider a mass M tied by a string of length R to the center with an angular velocity of ω radians per second. The mass rotates in a circular path because of the centripetal force,

$$F_{cf} = M\omega^2 R$$

CENTRIFUGAL SEDIMENTATION

The settling of colloidal particles.

Influence of *g* is not enough.

Say, a particle at *x* distance from centre.

Force acting on the particle

$\Rightarrow$ centrifugal force.

(towards the centre)

$= (m - m')\,\omega^2 x$

where , ω - angular velocity rad s^{-1}

$(m - m')$ is the apparent mass

force against the direction is the frictional force

$\therefore$ take a short –time to balance these forces

$(m - m')\,\omega^2 x = Bv(x)$

$= B\,dx/dt$

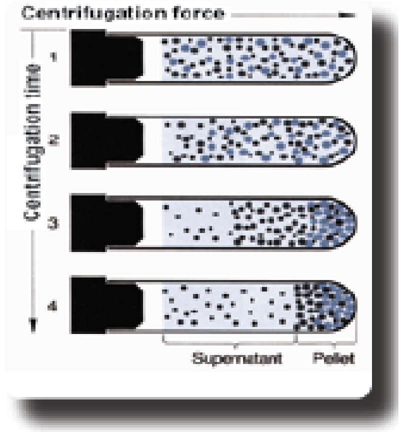


Diagram 2: Differential centrifugation of two substances with different sedimentation coefficients

As particle move towards end of the tube

v – increases

sendimentation coefficient,

$$S = \frac{v(x)}{\omega^2 x} = \frac{m - m'}{B}$$

Example:

Sendimentation.

$$(m - m') = \frac{1}{N_A}\left(M - \bar{v}_2 \rho_l\right)$$

$(m - m') = 1/N_A\ (M - \overline{V_2}\,\rho_1) = M\,(1 - \overline{v_2}\,\rho_1)\,/\,N_A$

$\overline{V_2}$ - partial molar volume of the aggregate (polymer)

$\overline{v_2}$ - volume per unit mass ($= \rho_s^{-1}$)

$\therefore$ $S = M\,(1 - \overline{v_2}\,\rho_1)\,/\,N_A B$

$= M D\,(1 - \overline{v_2}\,\rho_1)\,/\,RT$

$\therefore$ can estimate molar mass of the particle aggregate from S and D.

Use previous equation of $B\dfrac{dx}{dt}$

$$\frac{dx}{x} = \frac{M}{B}\left(1 - \rho_l / \rho_s\right)\omega^2 dt$$

$$= S\ \omega^2\ dt$$

After integration,

$$\ln x_2 / x_1 = S\omega^2 \left(t_2 - t_1\right)$$

From plot of $\ln x$ vs t

- s , ω = *normally known*
- apparent mass ($m - m'$) can be determined from

$$S = \frac{(m - m')}{B}$$

$$= \frac{(m - m')}{6\pi\eta r}$$

estimate , $B = \frac{kT}{D}$ (frictional coefficient from diffusion expt)

$$r = \left[\frac{9\eta S}{2(\rho_s - \rho_1)}\right]^{1/2}$$

Svedberg = 1S = 10^{-13}
Nobel Price 1926

Theodor Svedberg
(1884-1971)

Analytical Ultracentrifugation (AU)

An analytical ultracentrifuge spins a rotor at an accurately controlled speed and temperature. The concentration distribution of the sample is determined at known times using absorbance measurements.

The concentration, *c*, is determined for solutes obeying the Beer-Lambert law:

$$A = \varepsilon cl$$

where the absorbance, *A*, of the sample is measured at a given wavelength, ε, knowing the fixed position in the cell, *l*.

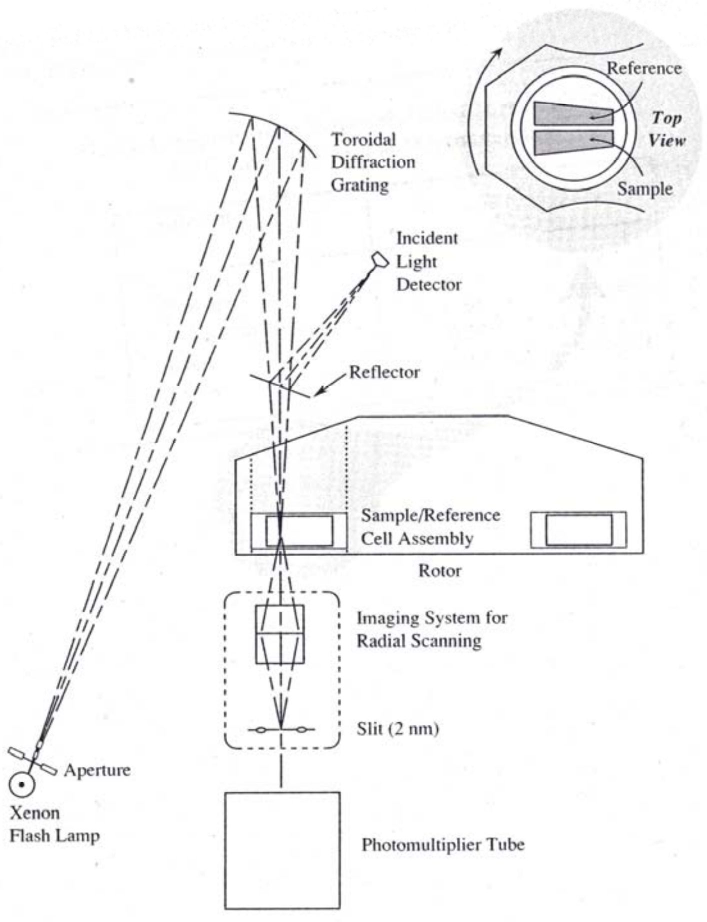
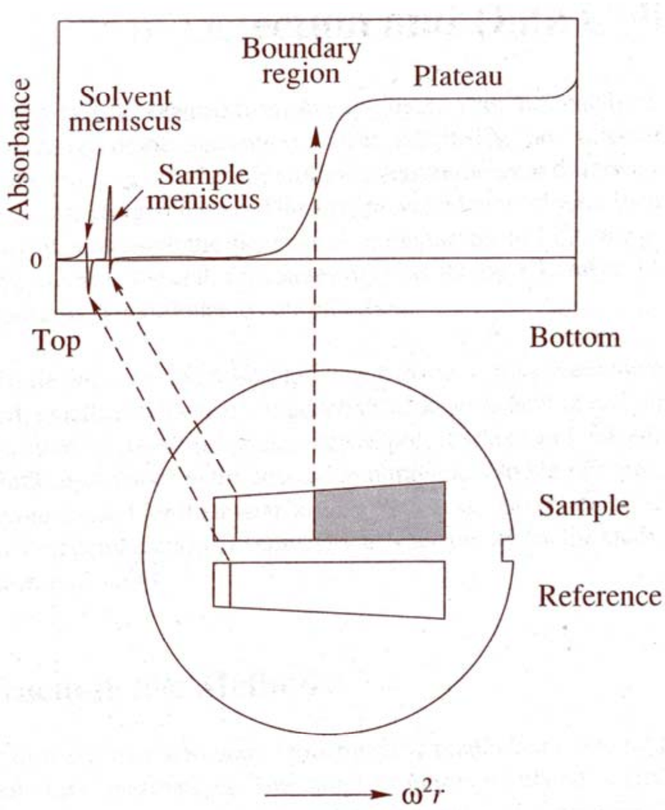
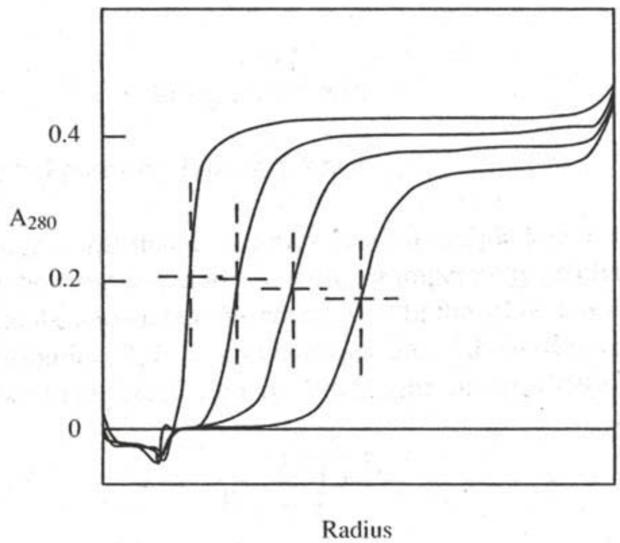


Figure: Beckman Optima XL-A absorbance system with a high intensity xenon flask lamp allows the use of wavelengths between 190 and 800nm.

For a sedimentation velocity experiment, an initially uniform solution is placed in a cell and a sufficiently high angular velocity is applied to cause rapid sedimentation of solute towards the cell bottom. As a result, there is a depletion of solute near the meniscus, causing a characteristic spectrum as shown in the following figure. A sharp boundary occurs between the depleted region and the sedimenting solute (the plateau).





Example:

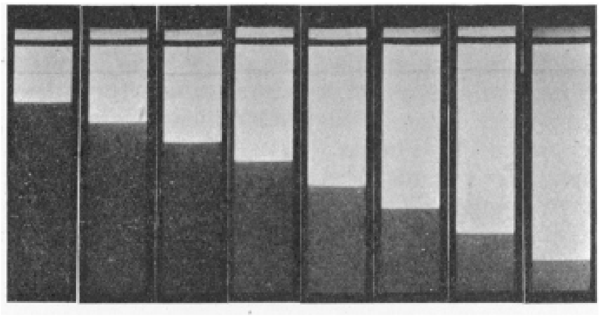


Fig. 1. A series of sedimentation pictures of the Bushy Stunt virus protein, 0·27 % in 0·02 *M* acetate buffer. Centrifugal force 20,000 g. 9 min. between each exposure.

In this case, $S = 146 \times 10^{-13}$ at 20 ^{o}C $\rho_v = 1.353$, r = 13.7μ and M= 8,800,000
http://www.biochemj.org/bj/032/1607/0321607.pdf

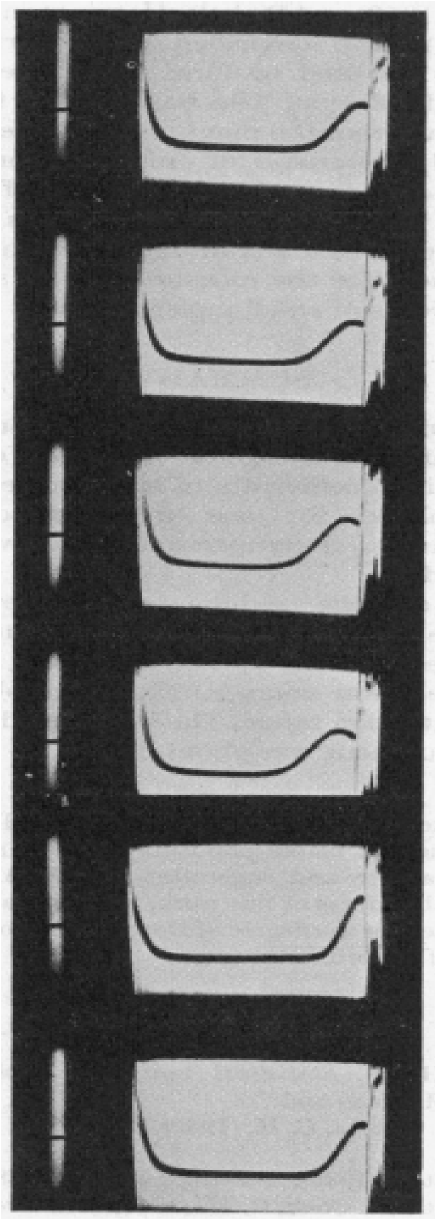


Fig. 2. Schlieren photographs showing the sedimentation of polyglucose A at concentration 1·40% (w/v). Each photograph represents a graph of refractive-index gradient versus distance in the cell. The fine vertical line near the right-hand side of each picture represents the meniscus between liquid paraffin and polyglucose solution. The time between photographs was 10 min., the speed of rotation 1000 rev./sec. The bar angle was 14·5° for the first four pictures and 18·5° for the last two.

TUTORIAL

Calculate the molecular weight of hemoglobin in water solution at 20°C, given that D = 6.3 × 10^{-7} cm^2 s^{-1}, V = 4.41× 10^{-13} s, v = 0.749 cm^3 g^{-1}, ρ = 0.9982 g cm^{-3}.
NB: 1 J = 1 Nm = Kg m^{-2} s^{-2} = 1000 g 10^{-4} cm^{-2} s^{-2}.= 0.1 g cm^{-2} s^{-2}.

$$s = \frac{MD}{RT}\left(1 - \bar{\upsilon}_2\,\rho\right)$$

$$\text{thus, } M = \frac{(8.314)(293)\left(4.41\times10^{-13}\right)}{\left(6.3\times10^{-11}\right)\left[1-\left(0.749\times10^{-3}\right)\left(0.9982\times10^{3}\right)\right]}$$

$$M = \frac{\left(JK^{-1}mol^{-1}\right)(K)(s)}{\left(m^2s^{-1}\right)\left[1-\left(m^3kg^{-1}\right)\left(kgm^{-3}\right)\right]}$$

M= 67.573 kg mol^{-1} = 67573 g mol^{-1}.

The Titanium dioxide pigment of density 4.12 g cm^{-3} is suspended in water at 33 °C. At this temperature the density and viscosity of water are 0.9947 g cm^{-3} and 7.523 × 10^{-3} Pa s, respectively. A light scattering measurement gives and average size of 0.29 μm. Given 1 Pa·s = 1 N· s m^{-2} = 1 kg m^{-1}·s^{-1}, J = kg m^2 s^{-2}. Calculate;

(a) Its diffusion coefficient.

$$D = \frac{kT}{6\pi\eta r} = \frac{1.38\times10^{-23}\times306}{6\pi\times7.523\times10^{-3}\times0.29\times10^{-6}}$$

$$D = \frac{4.223\times10^{-21}}{4.114\times10^{-8}} = 1.027\times10^{-13}m^2s^{-1}$$

(b) Sedimentation coefficient

$$r = \left[\frac{9\eta S}{2(\rho_S - \rho_l)}\right]^{1/2}$$

$$S = \frac{2r^2(\rho_S - \rho_l)}{9\eta} = \frac{2\times\left(0.29\times10^{-6}\right)^2\times\left(4.12\times10^{-3}\times10^{6} - 0.9947\times10^{-3}\times10^{6}\right)}{9\times7.523\times10^{-3}}$$

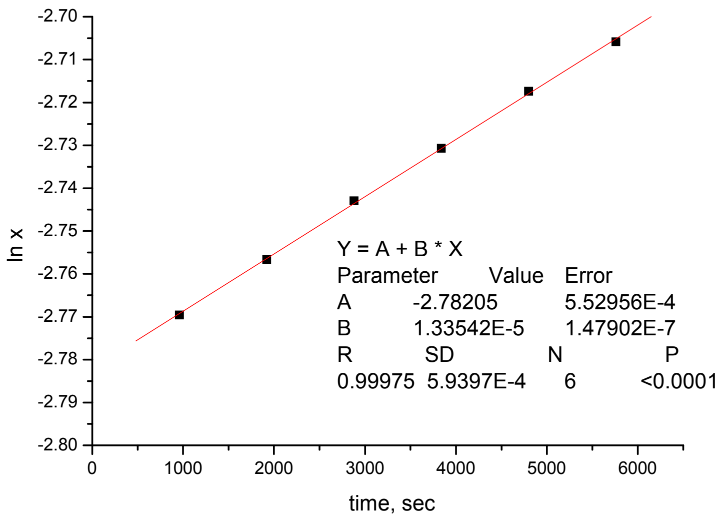
$$S = \frac{2\times8.41\times10^{-14}\times(3125.3)}{0.067707} = 7.764\times10^{-9}\,s$$

The sedimentation coefficient of a certain protein in 1.0 mol dm^{-3} NaCl at 20 ^{o}C was measured by boundary sedimentation at 24,630 rpm. The following data were recorded.

Time, (min)	Boundary distance, (cm)
16	6.2687
32	6.3507
48	6.4380
64	6.5174
80	6.6047
96	6.6814

Calculate the sedimentation coefficient *S* and the molar mass of the protein. The partial specific volume of sodium salt of protein is 0.556 cm^3 g^{-1}. The viscosity and density values of 1.0 mol dm^{-3} NaCl are 1.005×10^{-2} g s^{-1} cm^{-1} and 1.04 g cm^{-3} respectively.

Time, *t* (min)	Distance of boundary from center of rotation, *x* (cm)	Time, t (sec)	Distance, x (m)	Ln x
16	6.2687	960	0.062687	-2.7696
32	6.3507	1920	0.063507	-2.75661
48	6.4380	2880	0.064380	-2.74295
64	6.5174	3840	0.065174	-2.73069
80	6.6047	4800	0.066047	-2.71739
96	6.6814	5760	0.066814	-2.70584



$$\omega = (24{,}630\ \text{rpm})(2\pi)\left(\frac{1}{60}\right) rad\,\sec^{-1} = 2579.25\sec^{-1}$$

$$S = \frac{1}{\omega^2}(slope) = \frac{1}{(2579.25)^2}\left(1.335\times10^{-5}\right) = 2.01\times10^{-12}\ \sec$$

therefore, S = 20 S

Molar mass of protein, $M = \dfrac{SN_A\beta}{\left(1-\bar{v}\rho\right)} = \dfrac{SN_A(6\pi\eta r)}{\left(1-\bar{v}\rho\right)}$