THE MAGNETICALLY SUSPENDED EQUILIBRIUM ULTRACENTRIFUGE[1]

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An ultracentrifuge is described in which the rotor is magnetically suspended in a vacuum. The rotor is accelerated to operating speed by an air turbine below the vacuum chamber. A thin, flexible shaft passing through vacuum-tight oil glands connects the turbine to the rotor during the acceleration period but is disconnected during the operating period. The rotor coasts freely during the observation of the sedimentation. The rotor loses about 1 r.p.s. per 3 days when coasting freely in the vacuum chamber containing pressure of 10^{-6} mm. The sedimentation is observed with a modified type of Jamin interferometer. The rotor speed may be determined to one part in 10^6, the temperature to one part in 10^4, and the fringe shift to between 0.03 to 0.05 fringe. This gives from two to three significant figures in the precision over the entire molecular weight range above 100 when the equilibrium method is used.

Two principal methods are in general use for the measurement of molecular weights by centrifuging.[3] In the first, or rate-of-sedimentation, method the rate of settling of the substance in the centrifuge is determined, while in the second, or equilibrium, method the concentration of the substance is measured at various radial distances after equilibrium has been established between sedimentation and back diffusion. In both methods the rotor speed, the rotor temperature and the refractive index or its gradient are measured while the centrifuge rotor is spinning. The rate-of-sedimentation method employs a relatively larger centrifugal field but requires only a few hours for a molecular weight determination. On the other hand, the equilibrium method employs a relatively less intense centrifugal field but requires several days or weeks of continuous centrifuging for the determination. The theory of the rate-of-sedimentation method is based upon Stokes' law, while that of the equilibrium method is based upon thermodynamics and, therefore, is probably more reliable. Also, for the same rotor size and speed, much smaller molecular weights may be measured by the equilibrium method. Nevertheless, in the past where the molecular weights are large enough to give measurable rates of sedimentation in the centrifugal fields available, the rate-of-sedimentation method has been more widely used. The primary reason for this lack of popularity of the equilibrium method is the difficulty of maintaining the rotational speed and temperature of the centrifuge rotor sufficiently constant for long periods of time. The purpose of this paper is to describe a magnetically suspended vacuum-type ultracentrifuge which we are developing[4] principally for use with the equilibrium method, although it could equally well be used with the rate-of-sedimentation method. In this magnetically suspended centrifuge the absolute rotor temperature is maintained constant to at least one part in ten thousand; the troublesome "hunting" in the rotor speed is eliminated, and the rotor speed is measured to better than one part in a million. In the molecular weight measurements to be described it has been found advantageous to allow the rotor speed to decrease very slowly (the order of 1 r.p.s. per day) in order to reduce the centrifuging time.

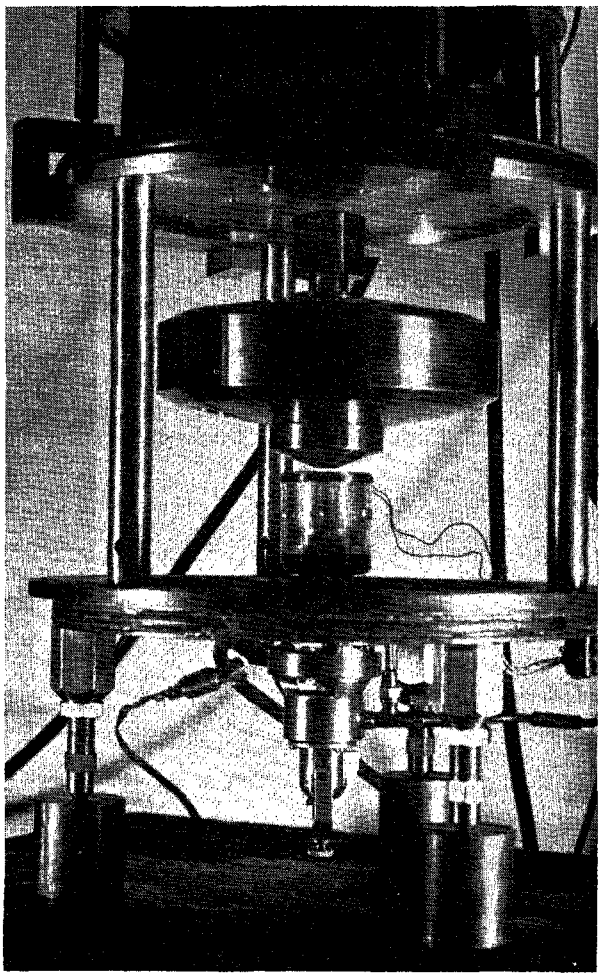

Fig. 1.—Photograph of ultracentrifuge with vacuum chamber removed.

Figure 1 shows a photograph of the apparatus with the vacuum chamber removed, and Fig. 2 is a cross-sectional diagram of the ultracentrifuge. The steel rotor is freely suspended inside of a nonmagnetic vacuum chamber by the support solenoid as indicated in Fig. 2. It is accelerated to operating speed by the air turbine situated below the vacuum chamber. A small-diameter, flexible steel shaft connects the air turbine with the rotor.

(1) Supported by Navy Bureau of Ordnance and National Science Foundation.

(2) (a) Union Carbide and Carbon Chemical Corporation fellow; (b) United States Rubber Company fellow.

(3) T. Svedberg and K. O. Pedersen, "The Ultracentrifuge," Oxford University Press, London, 1940.

(4) (a) J. W. Beams, J. D. Ross and J. F. Dillon, *Rev. Sci. Instru.*, **22**, 77 (1951); (b) J. W. Beams, *Science*, **120**, 619 (1954); *J. Wash. Acad. Sci.*, **37**, 221 (1947).

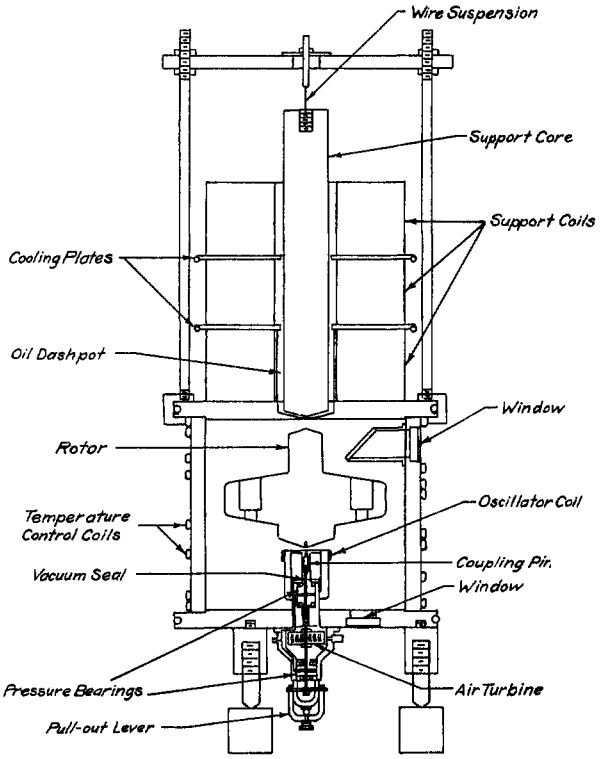

Fig. 2.—Diagram of ultracentrifuge.

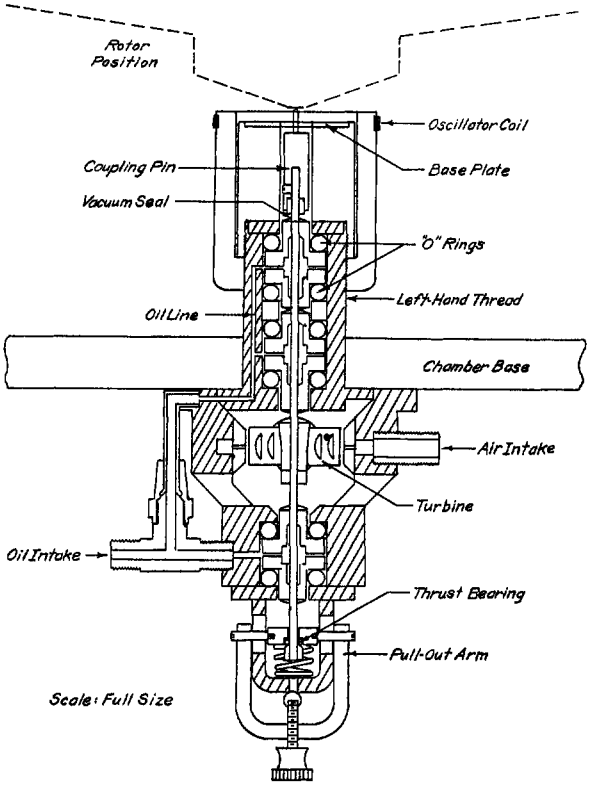

Fig. 3.—Compressed air drive system.

The shaft passes into the vacuum chamber through vacuum-tight oil glands as shown in detail in Fig. 3. During the acceleration period the shaft fits into a slot in the rotor; but, when the desired operating speed is reached, it is pulled out by a lever. This disconnects the rotor from the drive turbine and allows it to coast. The observations of the sedimentation are carried out while the rotor is coasting freely. The sedimentation is observed by passing light through the upper window, the right angle prism, down through a centrifuge cell (Fig. 6) mounted in the rotor, and out through the lower window.

The rotor shown in Figs. 1 and 2 was machined of one piece of high-strength alloy steel and weighs 13.5 kilograms. Its maximum radius is 9.4 cm., and its moment of inertia around the axis of rotation is 5×10^5 g. cm.^{2}. It has four 2.4 cm. holes drilled parallel to the axis of rotation with their centers 65 mm. from that axis. These holes carry the centrifuge cell shown in Figs. 4 and 6, a slit for chopping the light beam shown in Fig. 6, and two balancing cells. The centrifuge cell is similar to the matched double-sector type previously described.[4] Figure 4 shows a cross-sectional diagram of the cell. The cell consists of a housing internally threaded for the locking ring, washers, a double-sector spacer washer, two optically flat quartz windows with their optic axes perpendicular to their polished faces, separated by a spacer with two sectors as shown in Fig. 4. The housing and locking ring are made of duralumin ST 14, while the spacer which contains the double sectors between the windows is made of aluminum or duralumin covered with a thin coating when necessary to prevent corrosion. Grooves 0.004 cm. deep are machined into the upper and lower surfaces of this spacer in order to hold the 0.005 cm. polyethylene washers in place and seal the sector-shaped cells both from the outside and from each other. A keyway is milled into the inside wall of the housing which lines up the components of the cell and prevents them from twisting when the locking ring is tightened. Extreme care is taken in getting the

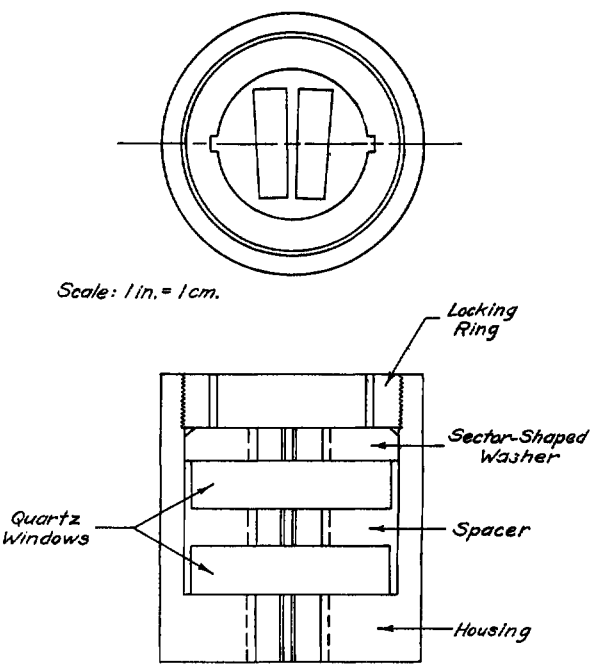

Fig. 4.—Double sector cell.

spacer flat; otherwise, leaks occur between the sectors, and the quartz windows are distorted by the pressure of the locking ring. Also, the sides of the sectors must be accurately flat. Radial lengths from 3 to 12 mm. and heights between 3 and 12 mm. have been used. The sector cells are filled, using a hypodermic syringe, through radial holes in the housing and spacers nearest the axis of rotation. These holes are sealed by threaded plugs with Teflon washers after the cells have been filled.

The cylindrical vacuum chamber is 22.2 cm. high, 23 cm. i.d., and a wall thickness of 1.76 cm. It is made of brass which is both non-magnetic and a good heat conductor. Copper cooling tubes are soldered to the vacuum chamber, and a heavy coating of glyptol is painted over the outside. The top and bottom plates of the vacuum chamber are 30 cm. in diameter and made from 1.9 cm. brass. They also have soldered copper cooling tubes (some of them in grooves for maintaining their temperature constant). Celvacene heavy vacuum wax is used to provide a vacuum seal between the end plates and the cylinder. Three heavy brass adjustable feet support the chamber and provide clearance for the air turbine assembly and parts of the optical system. Six glass windows 4.15 cm. in diameter and 1.02 cm. thick of good optical quality are arranged to provide three separate light paths through the rotor. The vacuum chamber is evacuated through a 7.6 cm. metal tube with flange connections to a Consolidated Vacuum Corporation V.M.F.-100 diffusion pump with the usual vapor traps and a "Megavac" fore pump. The pressure in the chamber is measured by a Pirani and an ionization gage. The temperature of the chamber was maintained constant to 0.03° by circulating water through the cooling coils and a thermostat bath in series.

The air turbine drive is shown in cross-section in Fig. 3. The duralumin turbine drives the rotor by a 0.25 cm. flexible steel shaft. The shaft turns in hard babbited bearings mounted in neoprene O-rings and lubricated with low vapor pressure oil ("Hy-Vac" oil freed of air and moisture). This type of bearing or gland has been described in detail previously[4] and is both slightly self-aligning and vacuum tight. The turbine is driven by compressed air admitted through small channels in the inner wall of the "air box" around the turbine. The air jets strike the turbine just above the center of the flutings and exert a slight upward force which automatically keeps the shaft engaged in the rotor while the air pressure (10 to 50 lb./in.^{2}) is applied. The brass housing is assembled in two sections. The upper section contains two bearings and extends up into the vacuum chamber. It is threaded to receive the two lucite pieces on which the oscillator coil and stainless steel base plate are mounted. Since the rotor turns counterclockwise when viewed from above, left-handed threads are used on the lucite. The base plate is of high-resistance non-magnetic 0.15 cm. stainless steel and protects the lucite if the rotor should get too low or in case of power failure. Recently Teflon has been found superior to lucite for the support of this base plate and for the base plate itself. A hardened steel "coupling pin" is attached to the upper end of the drive shaft with a set screw. The tip of the pin is hexagonally shaped to engage a modified Allen set screw located in the bottom of the rotor. A cylindrical neoprene gasket is fitted on the base of the coupling pin in such a way that, when the drive shaft is lowered to disengage the pin from the rotor, the gasket is squeezed against the top of the upper bearing and seals the vacuum chamber. This prevents oil from leaking into the vacuum chamber during the experiment. The lower section contains the turbine, lower bearing, and mechanism for disengaging the shaft from the rotor. Below the lower bearing a small knob is fastened to the end of the drive shaft and spins just below a ball-thrust bearing held up by a coil spring. The shaft is disengaged by pulling down on the pullout arm, and pressure is maintained on the chamber seal by tightening the lower thumb screw.

The support solenoid is wound in three sections or coils and contains a total of 36,000 turns of No. 22 insulated copper wire. Each section is approximately 24 cm. o.d. and 7 cm. i.d. and is separated by brass vanes cooled by water passing through copper coils soldered to the vanes. The soft iron core is 2.4 cm. diameter, 30 cm. long, and is suspended by a 0.0623 cm. piano wire inside the solenoid coil. The lower end of the core is a very flat cone (about 174°). The exact dimensions of this core are not critical. The lower end of the core is immersed in a dash pot of oil (No. 30 motor oil) and hangs about 0.15 cm. above the top of the centrifuge chamber. The top of the chamber is machined out to a thickness of 0.4 cm. just below the core as shown in Fig. 2. Any horizontal motion of the rotor is followed by the iron core, and, hence, quickly damped by the oil dash pot. The support circuit is shown in Fig. 5 and is similar to those used previously.[4a] It consists of a tuned-grid, tuned-plate oscillator operating at 3 megacycles followed by a detector from which an "error" signal and its time derivative are obtained. The time derivative serves as an anti-hunt or damping signal. The error and derivative signals are amplified separately and combined to control the current through a power stage whose load is the support solenoid. The oscillator or pickup coil is mounted on 5 cm. diameter lucite just under the rotor (Figs. 1 and 2) and consists of 10 turns of No. 22 copper wire. Leads are brought out of the chamber through Kovar seals and connected by a low-loss coaxial cable to the oscillator circuit. Vertical displacement of the rotor varies the impedance of the grid circuit which changes the amplitude of the oscillations and in turn the current through the solenoid. The current in the pickup coil is so small that no observable heating occurs. When properly adjusted, the rotor floats freely in the chamber without observable (100 power magnification) vertical or horizontal motion. In most of the experiments the rotor was supported approximately 2 mm. above the base plate and 3 mm. below the top of the chamber. To adjust the circuit, the rotor is placed upright on the base plate with the chamber removed to permit access

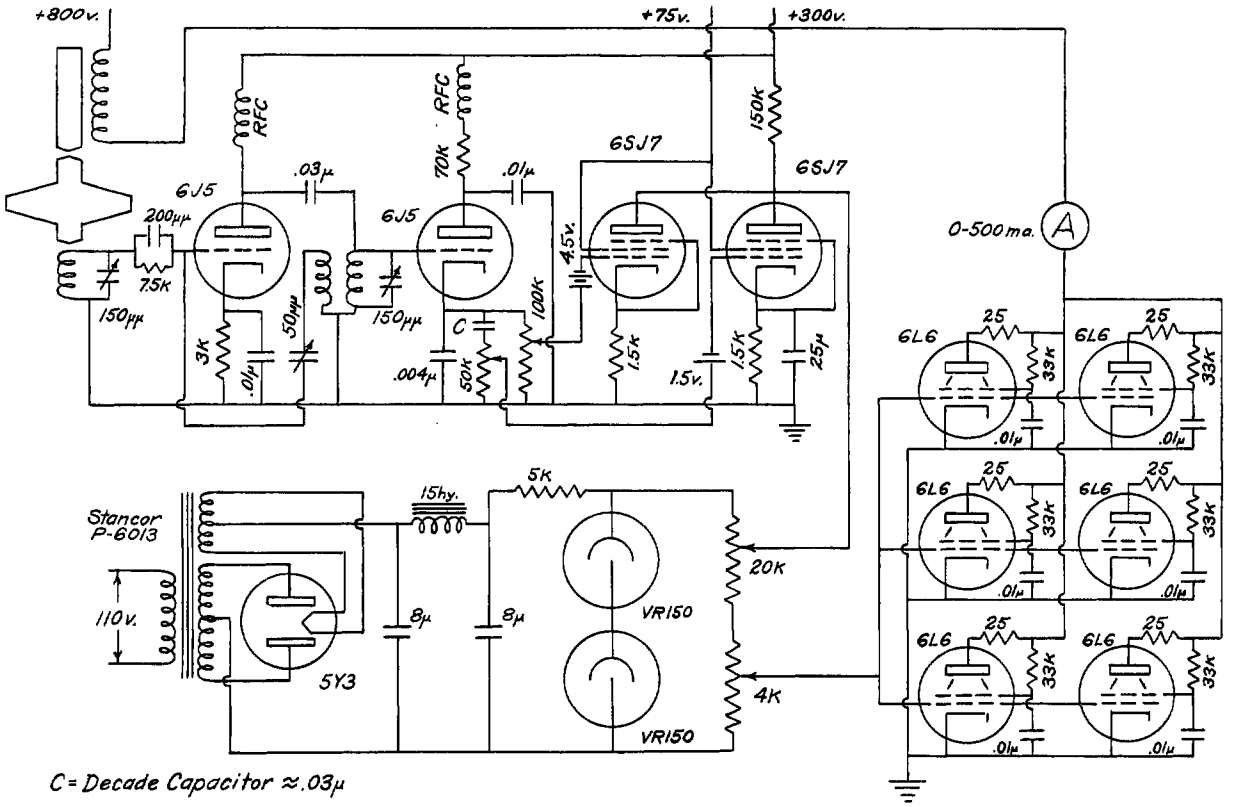

Fig. 5.—Rotor support circuit.

to the rotor. A high-resistance d.c. voltmeter is connected from cathode to ground in the 6J5 detector circuit and the grid and plate condensers tuned for a maximum output voltage (150 to 250 volts). The circuit is then neutralized to an output of about 50 volts. The rotor is next raised to the top of the chamber, and the voltage should increase to 150–200 volts. With the centrifuge completely assembled, the power supplies are turned on and the bias on the 6L6's adjusted until the rotor is picked up. The "error" potentiometer and the differentiating capacitor–potentiometer must next be adjusted until a stable support is obtained. When once adjusted for a given rotor, the circuit will remain in adjustment indefinitely. For the all steel rotor shown in Fig. 1 the support current is about 300 ma. Also the rotor friction is so small that the temperature of the rotor remains accurately equal to the surrounding vacuum chamber walls. When coasting freely at 300 r.p.s. with the vacuum chamber pressure of the order of 10^{-6} mm. of Hg, the rotor loses only about 1 r.p.s. per three days. As will be shown later, this gives sufficiently constant speed for the equilibrium experiments and, in fact, allows equilibrium to become established in a shorter time than if there were no deceleration. The rotor speed is measured by placing a photomultiplier tube in a light beam interrupted once for each rotation of the rotor (between M_2 and I_1 in Fig. 6). The resulting signal is then amplified and applied to the vertical plates of an oscilloscope. An amplified signal from the National Bureau of Standards radio station WWV or a signal from a generator calibrated and monitored with WWV is next applied to the horizontal plates of the oscilloscope. The two frequencies are compared by observation of the motion of the resulting Lissajous figures. The rotor frequency is adjusted to be near one of the broadcast frequencies

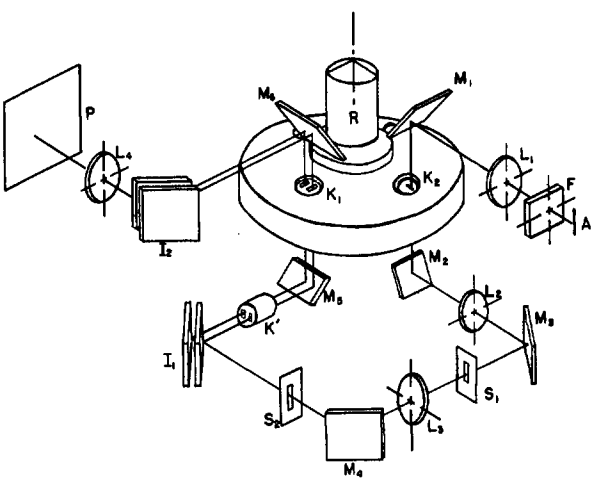

Fig. 6.—Combination light chopper and interferometer.

of WWV or one of its multiples or submultiples. The rotor speed easily may be measured to one part in 10^6. The high precision with which the temperature and the speed of the rotor can be controlled and measured makes it necessary to adapt more reliable methods to the determination of the index of refraction as a function of the rotor radius in the centrifuge cell. Fortunately, the develop-

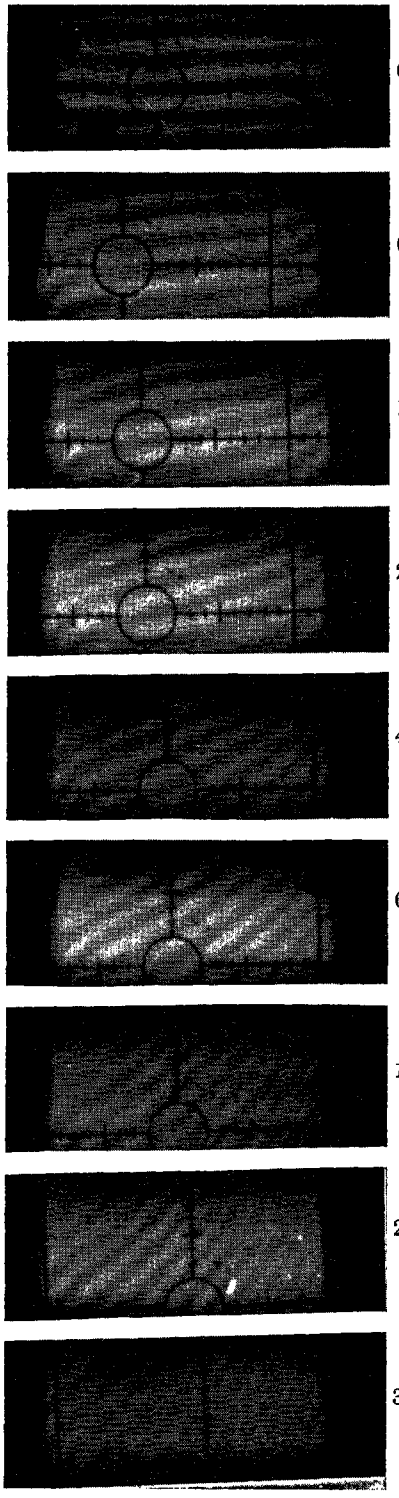

Fig. 7.—Photographs of fringes during experiment with human albumin.

ment of the double-sectored ultracentrifuge cell[5] makes it possible to use interferometer methods for these measurements. After several types of interferometer methods were tried, a modified Jamin (or Mach-Zehnder) type of interferometer was adapted to these measurements, because it not only gives precise values at various radial distances but also at all points throughout the ultracentrifuge cell (both parallel and perpendicular to the radius). The method has been described previously[6] in detail, so only a brief outline will be given. Figure 6 shows the combination light chopper and interferometer used. Light from a mercury arc is focused on the slit A and made monochromatic by the light filter F. The lens L_1 focuses the slit A on K_2 which is a radial slit in the centrifuge rotor so arranged that light can pass through only when K_2 is in the position shown. The light is next brought to focus on the slit S_1 and made parallel by the lens L_2. The width of the beam is limited by the diaphram S_2 to approximately the same as that of the sector-shaped cells in the centrifuge cell K_1. The light beam is then split by the interferometer I_1 and continues on through K^1 and K_1. The interferometer plates I_2 recombined the two beams, and the lens L_4 brings the cell K_2 to focus on the photographic plate P. The beam splitters I_1 and I_2 are each made of two optically flat glass plates. The front plate is lightly silvered so as to reflect from 25 to 35% of the light while the back plate is full silvered. If the plates in both I_1 and I_2 are accurately parallel, the interference fringes will be at infinity. However, if one of the plates in I_2 is rotated or inclined with respect to the other by a very small angle, the fringes are brought to focus on the photographic plate P. As a result both the image of K_1 and the fringes are brought sharply to focus on top of each other on P. The general theory of this type of interferometer has been given by Bennett and Kahl.[7]

The solution containing the substance to be analyzed is placed in one of the sector-shaped cells in K_1 and the solvent is placed in the other to precisely the same radial depth. The cell is then sealed and carefully aligned in the centrifuge rotor. The vacuum chamber is next evacuated to at least 10^{-5} mm. of Hg pressure, and the rotor with the drive shaft attached is magnetically supported. The rotor is then turned to the position shown in Fig. 6 by rotating the air turbine by hand. The interferometer is next adjusted so that one of the interferometer beams passes through the cell in K_1 containing the solution, while the other passes through the cell containing the solvent. Consequently, as sedimentation takes place in the solution, the interferometer fringes shift their position on the photographic plate P. The change in concentration ΔC due to a shift of one fringe is $\Delta C = \Delta n/k = \lambda/kh$ where n is the refractive index, λ is the wave length of the monochromatic light, k is the specific index of refraction increment, and h is the thickness of the cell K_1. It will be observed that, if one of the cells in the double cell K_1 is distorted with respect to the other by the effects of the centrifugal field, the fringes also will move. However, since the two cells have the same windows and housing, the effects of such distortions turn out to be negligible as shown by performing the experiments with the

(5) J. W. Beams and H. M. Dixon, III, *Rev. Sci. Instru.*, **24**, 228 (1953).

(6) J. W. Beams, N. Snidow, A. Robeson and H. M. Dixon, III, *ibid.*, **25**, 295 (1954).

(7) F. D. Bennett and G. D. Kahl, *J. Opt. Soc. Am.*, **43**, 71 (1953).

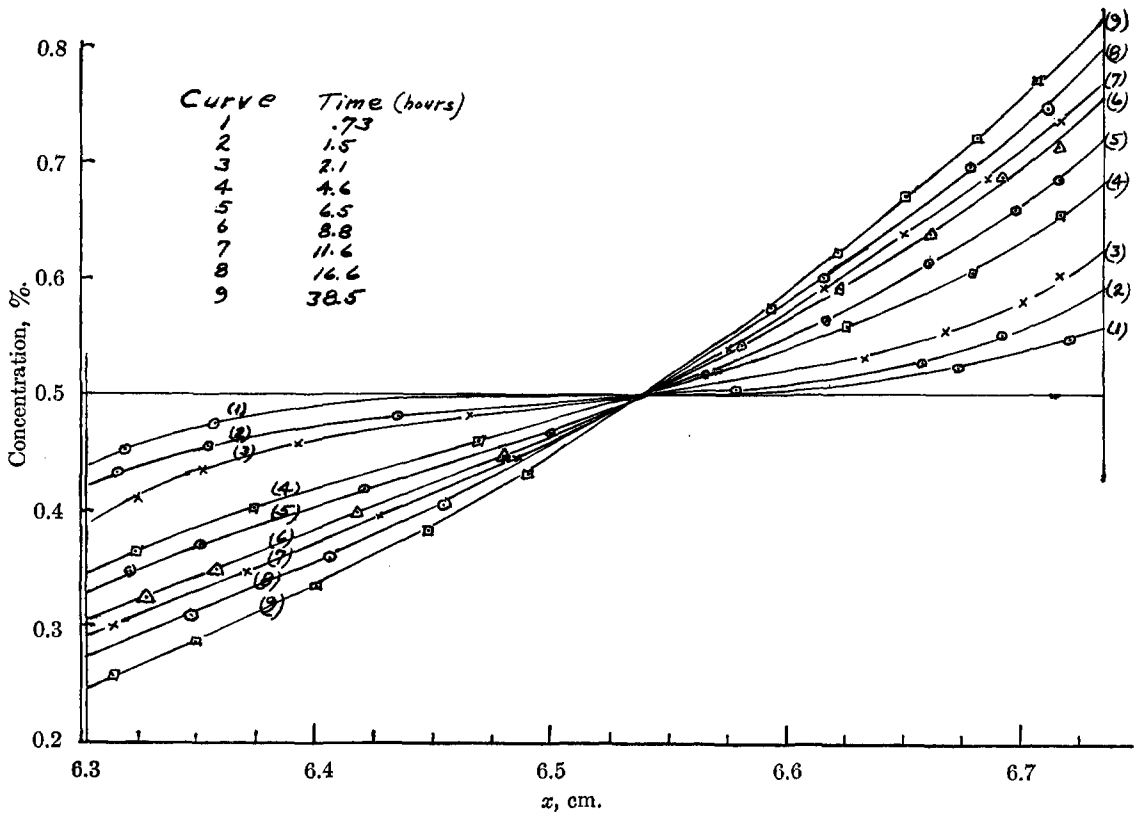

Fig. 8.—Distribution of concentration during albumin experiment.

solvent in both cells. In order to get white light fringes for reference, a cell K^1 identical to K_1 and filled in the same way is placed so that the beam which passes through the solvent in K_1 passes through the solution in K^1, while in the other beam the order is reversed. To obtain white light fringes, the filter F may be removed, or an incandescent light may be used as a source. At the beginning of the experiment white light fringes are obtained over the entire length of the cell (when they are parallel to the radius), but during sedimentation they occur only over a narrow radial region in the cell where the concentration in the sector of K_1 containing the solution is approximately the same as that at the beginning of the experiment. After the temperature of the rotor reaches a steady value, the fringes are adjusted so that they are either parallel or perpendicular to the radius of the centrifuge and are photographed in both monochromatic light and in white light. The centrifuge is then rapidly brought up to operating speed and the air turbine shaft disconnected. The fringes are again photographed both in monochromatic and white light at regular intervals until the observed concentration gradient in K_1 stops changing or until equilibrium is effectively established. As the rotor spins, light can pass the slit S_1 once each revolution and then only while the rotor turns through a small angle which is determined by the width of K_2, the properties and position of the lens L_2, and the width of S_2. In practice this angle is usually about 0.003 radian which gives sharp fringes over the entire image of K_1 on P. As a result precise values of the refractive index and, hence, the concentration are obtained for each point in the cell K_1 rather than at radial distances only.

For an ideal dilute incompressible solution, the molecular weight M_e of a mono-disperse substance obtained by the equilibrium method is given by the relation[3]

$$M_e = \frac{2RT \log_e (C_1/C_2)}{(1 - dV)4\pi^2N^2(X_1^2 - X_2^2)}$$

where C_1 and C_2 are the concentrations at the radial distances X_1 and X_2, respectively, d is the density of the solution, V the partial specific volume, T the absolute temperature, and N the number of revolutions per second. When the solution is not ideal and dilute, the above relation must be corrected[3] as shown especially by Williams and his students.[8] In any case, the three quantities to be determined, while the centrifuge is spinning, are the rotor temperature, the rotor speed, and the concentration at various radial distances in the cell. This is also true for molecular weight distribution measurements of polydisperse substances. The other factors, such as V and d, are determined outside the centrifuge.

Figure 7 shows a series of photographs of fringes taken with monochromatic light at various times during the progress of the sedimentation of human serum albumin. Photographs, not shown in Fig. 7, taken up to 65 hours showed only a very slight change in the fringes after about 32 hours. The solution at the beginning was 0.5% albumin in 0.01 N NaCl with sodium acetyltryptophan

(8) J. W. Williams, *J. Polymer Sci.*, **12**, 351 (1954).

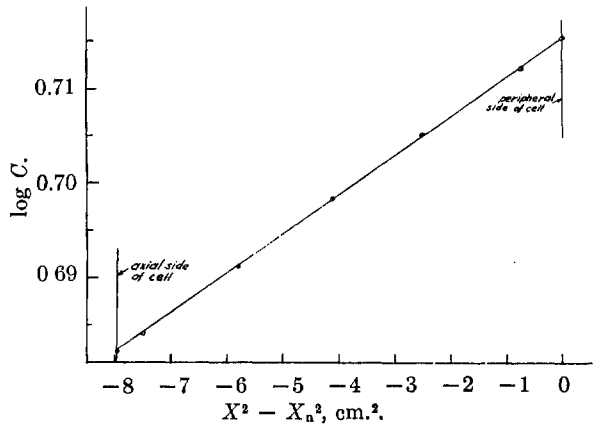

Fig. 9.—Log C *versus* $X^2 - X_n^2$ for sucrose after 58 hours centrifuging.

buffer. The rotor speed at the beginning of the experiment was 130.7 r.p.s. and at the end 129.9 r.p.s. Figure 8 shows the measured concentrations plotted as a function of the radial distance X as equilibrium is approached in a single experiment. Assuming the material to be monodisperse, the molecular weight values were found to depend upon the source, the past treatment and apparent purity of the human albumin used. Figure 9 shows the log C at radius X plotted *versus* $X^2 - X_n^2$, where X_n is the radius of the periphery of the cell for sucrose dissolved in water. The rotor speed was close to 300 r.p.s. According to theory at equilibrium, this should be a straight line, the slope of which gives the molecular weight. The value obtained was 343, which agrees very well with the known value of 342.3. The curve of Fig. 9 gives the values after 58 hours of centrifuging sucrose, but after 12.5 hours the data all fall upon a straight line. All of the data taken on a number of different substances indicate that equilibrium is approached much faster than should be expected from the well known formula.[8] This results, in part at least, from the fact that the rotor speed is very slowly decreasing. Archibald[9] has shown that, if $(1/N)\ (dN/dt)/4\pi^2N^2S << 1$, where S is the sedimentation constant and N the rotor speed, then the equilibrium condition will remain once it is established in the cell.

Since the precision of the rotor speed may be obtained to about one part in 10^6 and that of the temperature to roughly one part in 10^4, the method of measuring the concentration as a function of the radius is still the factor which limits the accuracy of the results obtained at least during the centrifuging process. In practice at present a fringe shift of between 0.03 and 0.05 is about the best that can be measured with reliability. This gives a precision of about three significant figures for substances with molecular weights from about 10^2 up to the largest values known. It is usually possible to work with both relatively low or high concentrations of a substance, since the total fringe shift (and, hence, the accuracy) is increased by increasing the rotor speed. Fortunately the rotor speeds required usually are well below those obtainable with the apparatus. Nevertheless, it is highly desirable especially for measurements in extremely dilute solutions to have a more precise method of determining the concentrations in the centrifuge cell. It is well known, of course, that greater precision should be obtained if a multiple beam interferometer could be used. One of us in collaboration with Mr. D. R. Carpenter is attempting to develop this technique for the ultracentrifuge, but the results are as yet only of a preliminary nature.

We believe the centrifuge described in this paper provides an excellent equilibrium method of getting molecular weights, since it gives good precision and the data are obtained in a relatively short time. Also, it is about equally useful over the entire range of molecular weights above 100. However, there are several rather obvious improvements which we have under development that should make the apparatus more widely adaptable. For example, it is of much interest to investigate the methods of obtaining molecular weights by measuring the approach to equilibrium as suggested by Archibald.[10] This requires a very rapid acceleration of the rotor followed by an extremely constant speed. The air turbine drive described above gives the rapid acceleration without rotor heating (actually, the over-all rotor cools slightly because of stretching) but does not give the constant speed required. For some time we have been developing an electrical drive in which the rotor is essentially the armature of a synchronous motor. In order to avoid excessive heating of the rotor during the period of acceleration, the drive frequency is increased automatically at the same rate as the rotor speed. As soon as operating speed is reached, the drive frequency is controlled by a piezoelectric crystal circuit which maintains the speed contant[11] to at least one part in 10^7. The principal problem is to produce rapid acceleration without excessive rotor heating.

DISCUSSION

J. MYER (*Communicated*).—Between 1951 and 1953 a magnetically suspended centrifuge was being constructed at the University of Southern California in collaboration with Prof. K. J. Mysels, based on the earlier description (*Rev. Sci. Inst.*, **23**, 77 (1951)) given by Prof. Beams. In this connection we had to develop independently many of the improvements responsible for the good functioning of the suspension mechanism now described. There are only a few other developments worth mentioning in addition to those presented in Prof. Beams' paper:

(1) The efficiency of the magnet is greatly increased if the magnetic lines of force are guided in ferromagnetic material, not only in the core, but also above and down around the outside of the solenoid. This is achieved by suspending from the piano wire a rigid assembly of a core (of ARMCO iron), an end-plate and a coaxial cylinder surrounding the coils. (2) A further increase in lifting force is obtained by making the top of the rotor in the shape of a quite flat mushroom of ARMCO iron so that most of the lines of force are confined to the region above the body of the rotor. (3) The rotor and inside of the vacuum chamber are blackened to insure efficient radiative heat transfer. (4) The plate between the magnet and rotor supports the whole assembly and is mounted permanently on an elevator. After adjustment the assembly is lowered into the vacuum chamber (for which the plate forms a lid sealed by one O ring). (5) On the circuit side we wind the sensing coil with silver wire on glass

(9) W. J. Archibald, private communication.

(10) W. J. Archibald, THIS JOURNAL, **51**, 1204 (1947).

(11) J. W. Beams, E. C. Smith and J. M. Watkins, *J. Soc. Motion Pict. and Tele. Eng.*, **58**, 159 (1952).

for easier degassing and to improve the Q of this coil and hence the sensitivity. 5881 tubes give better power handling and reliability than 6L6's. To obtain stability, the oscillating circuit is well protected from the heat generated in the rest of the circuit. (6) Our drive mechanism (of very low power) is based on a hysteresis motor. Water-cooled field coils within the vacuum chamber drive a short sleeve of hysteresis alloy mounted on the "stem" of the mushroom above the body of the rotor.

J. W. BEAMS.—First I wish to congratulate Dr. Meyer and Dr. Mysels on their very nice magnetically supported centrifuge. It illustrates the importance of parallel development. It might be of interest to mention that Dr. A. R. Kuhlthan in our laboratory has in some unpublished work developed a reasonably precise radiation method of measuring rotor temperatures and finds that if appreciable heat is developed in the rotor blackening of the rotor and chamber walls is helpful. In our experiments the rotors and walls also were made effectively black although so little heat is generated in the rotor that this is not necessary. Incidentally if there is small comparatively short time variations in the wall or chamber temperature, as often occurs, in some thermostatic devices, it is better to make the rotor bright. The rotor temperature will then not vary as much as that of the chamber, if the vacuum is good. Also we have had some experience with guiding the magnetic flux by placing additional iron around the solenoid. In general it seems to reduce the ease of regulation. On the other hand it reduces the stray flux. The power required does not seem to be excessive without it. We have suspended masses which varied in weight from 10^{-6} g. to 5×10^{4} g. with different solenoids.

B. JIRGENSONS.—What values of molecular weight have been arrived at for such materials as serum albumins by this method? Have you studied albumins from different sources?

J. W. BEAMS.—Albumin from different sources seems to have different degrees of homogeneity. Assuming homogeneity some samples gave values as low as 60,000 while others gave values of above 70,000. A specially purified sample kindly supplied by Dr. H. Hoch gave a value of approximately 74,000.

G. KEGELES.—Is it at all serious that there is an adiabatic cooling of the rotor upon acceleration, due to stretching?

J. W. BEAMS.—There is, of course, a slight cooling of the rotor when the rotor is accelerated to operating speed. However this is very small in the equilibrium centrifuge because the rotor speed is relatively small. In any case the rotor temperature approaches very closely to that of the walls in a time which is negligible in comparison to the time for equilibrium to take place.

AN AIR-DRIVEN, AIR-FLOATED CAPILLARY TUBE ULTRACENTRIFUGE[1,2]

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An air-driven, air-floated ultracentrifuge of the Henroit and Huguenard type is described. The rotor is constructed with opposed radial holes in which glass capillary cells are inserted. The rotor may be stopped at appropriate intervals during the course of centrifugation, and the tubes removed and photographed. A simple optical system utilizing a horizontally mounted research microscope is used to obtain scattered light, absorption and schlieren pictures. Sedimentation velocity results are reported on earthworm blood, snail blood, and tobacco mosaic virus protein. The agreement with results reported, using other ultracentrifuges, is good. For tobacco mosaic virus certain exaggerated dilution effects at low concentrations are noted, probably attributable to the use of non-radial cells. This is not serious and may be taken into account. Although the rotor is run in air, the temperature at the position of the cell is essentially the same as the room temperature, and effects due to temperature gradients are negligible. Diffusion constants can be calculated from photographic schlieren records, of the spreading of centrifugally produced concentration gradients, made while the tube is mounted vertically on the microscope stage. Asymptotic packing volumes of the sedimented erythrocruorin of worm blood and of the colloidal proteins of milk can be measured from photographs of the cells after centrifugation at increasing centrifugal forces. The packing volumes agree satisfactorily with the voluminosities calculated from viscosity measurements using the Einstein equation. They also agree, for the colloidal milk proteins, with voluminosities calculated from analyses of deposits compacted in bowl rotors.

Introduction

The ultracentrifuge design and technique presented are based on the fact that liquid samples confined in sufficiently small cells are so completely immobilized that the centrifuge rotor can be repeatedly stopped and the cells removed for photographing without sensibly altering the sedimentation gradients. Cylindrical cells made from ordinary glass capillary tubing are used. The technique is simple and permits the ready measurement of sedimentation constants, diffusion constants, and voluminosities, all, if desired, on a single sample. Results for erythrocruorin of earthworm blood, hemocyanin of snail blood, and tobacco mosaic virus protein are here reported.

Immobilization of liquids during centrifugation by using capillary cells seems to have been described first by Elford.[3] Most of his cells were of metal. The progress of sedimentation was followed by bacteriological and chemical analyses of the contents of the cells after centrifuging for different times. Adaptations of this basic Elford analytical technique have been described by McIntosh and Selbie,[4] who, however, used cells actually of more than capillary diameter; by Polson[5]; by Ford and Ramsdell[6]; and by Brakke, Block and Wyckoff.[7] The last-named authors used long glass capillaries,

(1) Presented at the McBain Memorial Symposium, Colloid Section of the American Chemical Society, Chicago, September, 1953. The experiments on tobacco mosaic virus have, however, been added since.

(2) The work here described was done in the U. S. Department of Agriculture and supported in part by Bankhead-Jones Special Research Funds. Preliminary work was done previously by the senior author in the laboratory of the Shell Development Company, Emeryville, California.

(3) W. J. Elford, *Brit. J. Exp. Path.*, **17**, 399, 422 (1936).

(4) J. McIntosh and F. R. Selbie, *ibid.*, **18**, 162 (1937).

(5) A. Polson, *Nature*, **148**, 593 (1941).

(6) T. F. Ford and G. A. Ramsdell, "XIIth International Dairy Congress, Section II, Subject 1," Stockholm, 1950, p. 17.

(7) M. K. Brakke, L. M. Block and R. W. G. Wyckoff, *Am. J. Botany*, **38**, 332 (1951).