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Constant speed drive for the magnetically supported equilibrium ultracentrifuge*

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A constant speed drive (10^{-3}%) is described for the magnetically supported equilibrium ultracentrifuge

In the equilibrium ultracentrifuge method of measuring molecular weights, two rigid requirements must be met. First, no material in the solution can be allowed to deposit on the peripheral cell wall, and second, equilibrium must be finally established between sedimentation and diffusion, or it must be possible to find this equilibrium by unambiguous extrapolation. In the case of solutions of large macromolecules such as the viruses, the first restriction imposes a relatively low limiting speed on the ultracentrifuge rotor, while the second requires that the sedimenting column be completely free of convection. Unfortunately, with low rotor speeds the sedimenting column becomes extremely sensitive to temperature gradients, variations in rotor speed, etc. which generate convection. The magnetically supported ultracentrifuge, previously described, in which the rotor coasts in a vacuum during the experiments, has been found to meet the above requirements.[1] However, in the case of the large molecules ($>10^6$ daltons) it is usually necessary to carry out a tedious extrapolation to the equilibrium state from quasiequilibrium states, since the rotor speed ($\sim$10 rps) is slowly decreasing. This paper describes a constant speed drive ($\pm 10^{-3}$% precision or $\pm 10^{-4}$ rps at a speed of 10 rps), for the ultracentrifuge rotor, which is essentially free of hunting. The drive can also accelerate or decelerate the rotor at about 1 rps/min without appreciable heating of the rotor. Consequently, the external drive used previously, with its accompaning oil seals, can be abandoned, which in turn eliminates all contamination of the optical surfaces.

The drive torque is provided by the system of Fig. 1. Two small ceramic magnets mounted in the base of the rotor rotate between a pair of fixed stator coils, in which current is commutated by an optical pickup. When light is reflected by the polished half of the rotor's rim, current flows in one direction; when light is absorbed by the black portion of the rim, however, the current flows in the opposite direction. The direction of torque is selected by choosing the Q or $\bar{Q}$ output of the Schmitt trigger, and the torque magnitude is determined by the power supply voltage. The current reversing switch is equivalent to a DPDT relay connected to reverse the polarity when energized by the dc amplifier.

The control switch couples the power supply to the reversing switch only when a logical one signal is provided, which permits control of the rotor speed.

The rotor is held at nearly constant speed by the circuit of Fig. 2, which supplies a logical one signal to the drive circuit control switch when the speed is too low. Once each second, the circuit determines whether or not the speed is sufficient. This is done by comparing a multiple of the period of rotation to a one-second time interval precisely generated by a reference oscillator. In the circuit shown, two counters are synchronized (by the INHIBIT and SHIFT flip–flops) to begin counting at the leading edge of a pulse from the

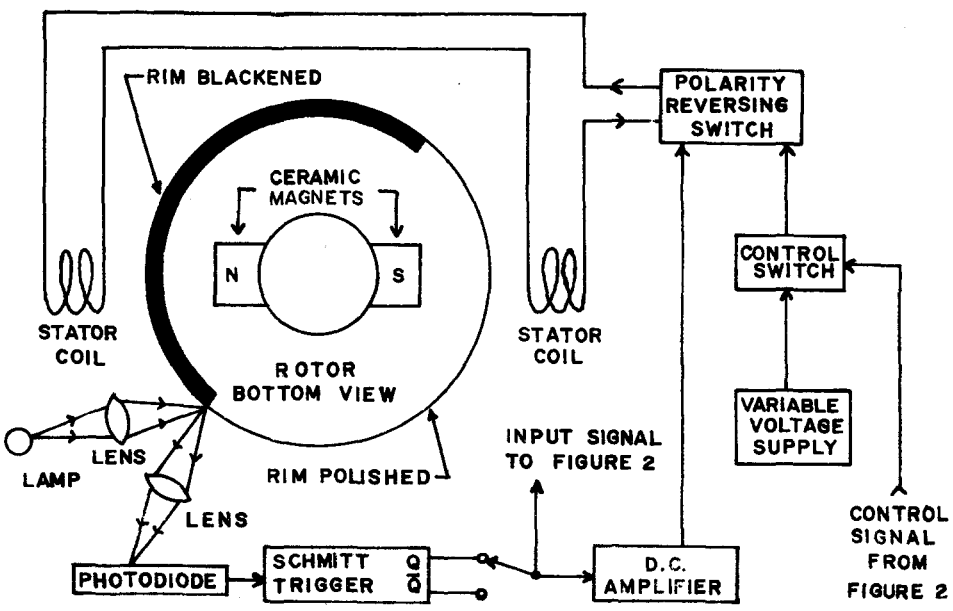

FIG. 1. System providing drive torque.

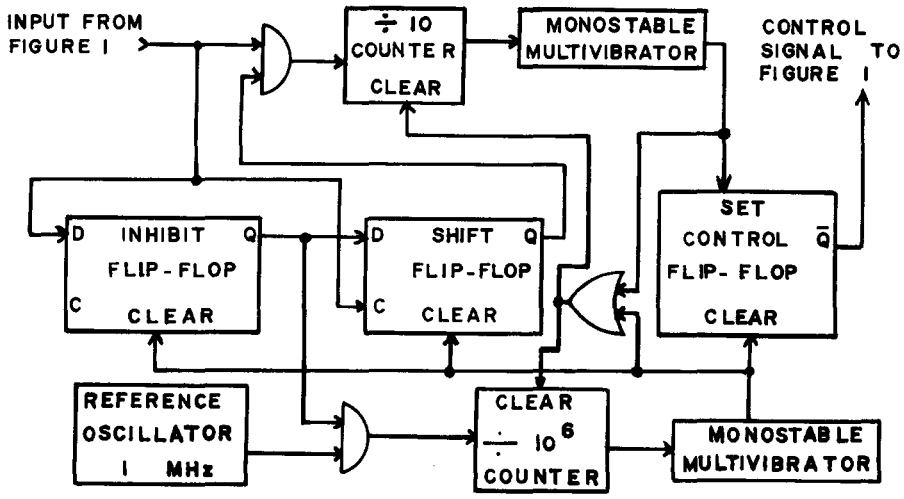

FIG. 2. Speed control circuit with rotor held at nearly constant speed.

Schmitt trigger of Fig. 1. If the rotor speed exceeds 10 rps, the upper counter triggers the upper monostable multivibrator, which sets the control flip and clears both counters. A new determination cycle begins immediately, and no drive is provided (the $\bar{Q}$ output of the control flip–flop is at logical zero). However, if the rotor speed is below 10 rps, the lower counter triggers the lower monostable, which clears the control flip–flop, both counters, and the INHIBIT and SHIFT flip–flops. The logical one $\bar{Q}$ output of the control flip–flop activates the drive system during the succeeding determination period, which begins at the next leading pulse edge from the Schmitt trigger.

Synchronization at the leading edge of the trigger pulse renders the precision independent of the rotor speed, an important advantage with the low speeds required for the ultracentrifuge. When the rotor is at the proper speed, each determination period begins and ends at a particular orientation of the rotor. Since the inherent deceleration of the coasting rotor is less than 1% per day, the theoretical limit of speed control with this instrument is $\pm 10^{-5}\%$ with a one-second sampling interval. Using a precision ($\pm 10^{-6}\%$) reference oscillator, and T^2L circuit components, we have routinely been able to maintain the rotor speed constant to $\pm 10^{-3}\%$ in week-long sedimentation equilibrium experiments.

This apparatus has been used for the study of heavy monodisperse plant viruses. For example, the molecular weight of turnip yellow mosaic virus was found to be 5.55×10^6 daltons, and its second virial coefficient was approximately 3×10^{-6} ml-mol/g^2. This nonideality exceeds the excluded volume effect; the excess has been attributed to the Donnan effect resulting from a large macromolecular charge, partially compensated by preferential interaction with the supporting KCl electrolyte.[2]

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[1]J. W. Beams, R. D. Boyle, and P. E. Hexner, Rev. Sci. Instrum. 32, 645 (1961).
[2]J. H. McGee, D. W. Kupke, W. Godschalk, and J. W. Beams Proc. Natl. Acad. Sci. (to be published).

Precise calibration method for time-to-amplitude converter*

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Accurate calibration of systems using time-to-amplitude converters can be difficult and time consuming. A simple and quick method for calibrating such systems is described. The method has wide operating range and high precision and uses equipment which is commonly available in the laboratory.

Precise calibration of a time-to-amplitude converter (TAC)–multichannel analyzer system requires two identical pulses separated by an accurately determined time interval. In the most common method of calibration[1] a fast pulse is split into two branches; one pulse goes directly to the START input of the TAC and the second travels through a calibrated length

