Reprinted from THE REVIEW OF SCIENTIFIC INSTRUMENTS, Vol. 3
Printed in U. S. A.

Apparatus for the Measurement of Optical Rotation of Solutions at High Pressure*

S. J. GILL AND ROBERT L. GLOGOVSKY
Department of Chemistry, University of Colorado, Boulder, Colorado

(Received 4 March 1964; and in final form, 1 June 1964)

A polarimeter was constructed so that the sensitive optical components could be subjected to the same pressure within a high pressure optical bomb, thereby avoiding the problem of large double refraction effects from two windows under a large difference of pressure. One of two Polaroids within the high pressure bomb is rotated through a fixed angle by a mechanical coupling with a solenoid driven magnet. The transmission of light for a given wavelength setting is measured in both Polaroid positions by means of a Beckman DU spectrophotometer. The sensitivity of the apparatus is $\pm 0.01°$ in the range of 0–5° rotation. The apparatus has been specifically designed for pressure studies to 2000 atm, although the method is inherently capable of even higher pressures.

INTRODUCTION

An interest in studying the thermodynamics of helix-coil transitions has led us to investigate the possibility of measuring the optical rotation of solutions under high pressure conditions. Siertsema[1,2] and others[3,4] have made attempts to measure optical rotation under high pressures, but the accuracy of the results has been impaired by the effect of double refraction in the windows of the high pressure cell.

Some recent polarimeter attachments[5] for spectrophotometers make use of a principle which enables one to eliminate the complication of the birefringent effects of the bomb windows. The optical rotation is detected by two nearly crossed polarizing elements, one of which can easily be rotated at a fixed angle. The intensity of light passing through this system in the two extreme positions can be nearly equalized by proper adjustment. If an optically active material is placed between the two polarizing elements, then a rotation of light occurs so that the intensities of light pass through the system will be different for the two fixed positions of the polarizer.

The ratio I/I_0 of the emergent to incident beam intensity is dependent upon the angular orientation of the polarizer with respect to the analyzer and upon the amount of optical rotation introduced between the two polarizing elements.

As one intuitively expects, a Stokes matrix analysis[6–8] of the system shows that the birefringence of the bomb windows has no influence on the intensity ratio. The problem of absorption and reflection losses is avoided by making two measurements on I, one differing from the other by a change in the polarizer orientation. One then finds that the transmission T, defined by the ratio of emergent intensities for angles of uncrossing θ_1 and θ_2, that

$$T=I_1/I_2=\sin^2(\theta_1-\alpha)/\sin^2(\theta_2-\alpha), \qquad (1)$$

where α is the optical rotation of the region between the Polaroids. Gallop[9] has derived a simplified formula for the case where $\theta_1=-\theta_2$. This condition is tedious to achieve in the apparatus we have constructed, but a simple calibration can be made to provide a measurement of the angular deviation β. Then $\theta_1+\beta=-\theta_2$. Gallop's result then becomes

$$\alpha=-\tfrac{1}{2}\beta+(\theta_1+\tfrac{1}{2}\beta)\tanh(0.576D). \qquad (2)$$

The determination of α and θ_1 is made by measuring D for one or more solutions of known α. When D is less than 0.2 for our purposes, the hyperbolic tangents can be approximated by the argument.

APPARATUS

The polarimeter apparatus, which fits inside the high pressure bomb,[10] is shown in Fig. 1. The solution cell is made of Teflon and the ends of the cell accommodate 1-cm-diam polarimeter windows.

Slotted spacing washers are placed at the ends of the Teflon cell to provide complete flow of the pressure fluid around the cell. Two springs (one partially shown) are held in slots in the housing and clip into the spring washers on the right. These springs seat the cell against the rotatable polarizer assembly which is held to the housing by set screws.

The polarizer assembly, consisting of two bearings, cam, cam stop disk, and polarizer, is also shown in Fig. 1. The cam can be rotated a precise amount determined by the cam stop disk. The Polaroid material used has the transmission characteristics corresponding to HN-32.[11] The cam

* This work was supported by grants from the Research Corporation, the Institute of Public Health, Department of Health (C-5393), and the National Science Foundation (GP-734).

[1] L. H. Siertsema, Rept. Amsterdam Acad. Sci. **5**, 305 (1896–97).
[2] L. H. Siertsema, Arch. Neerl. **3**, 79 (1900).
[3] F. V. Sander, J. Biol. Chem. **148**, 311 (1943).
[4] R. H. Zinszer, Trans. Kansas Acad. Sci. **41**, 241 (1938).
[5] Two commercial instruments, the Keston unit, manufactured by the Standard Polarimeter Company, New York, and a prototype model by Perkin-Elmer Corporation, employ this principle. A description is given by C. Djerassi, *Optical Rotatory Dispersion* (McGraw-Hill Book Company, Inc., New York, 1960), p. 32.
[6] W. H. McMaster, Rev. Mod. Phys. **33**, 8 (1961).
[7] M. J. Walker, Am. J. Phys. **22**, 170 (1954).
[8] W. A. Shurcliff, *Polarized Light* (Harvard University Press, Cambridge, Massachusetts, 1962), Appendix 2.
[9] Paul M. Gallop, Rev. Sci. Instr. **28**, 209 (1957).
[10] S. J. Gill and W. Rummel, Rev. Sci. Instr. **32**, 752 (1961).
[11] We found that this Polaroid material was not influenced significantly by application of pressure. Some initial results with glass-laminated polarizer disks gave erratic behavior to pressure changes.

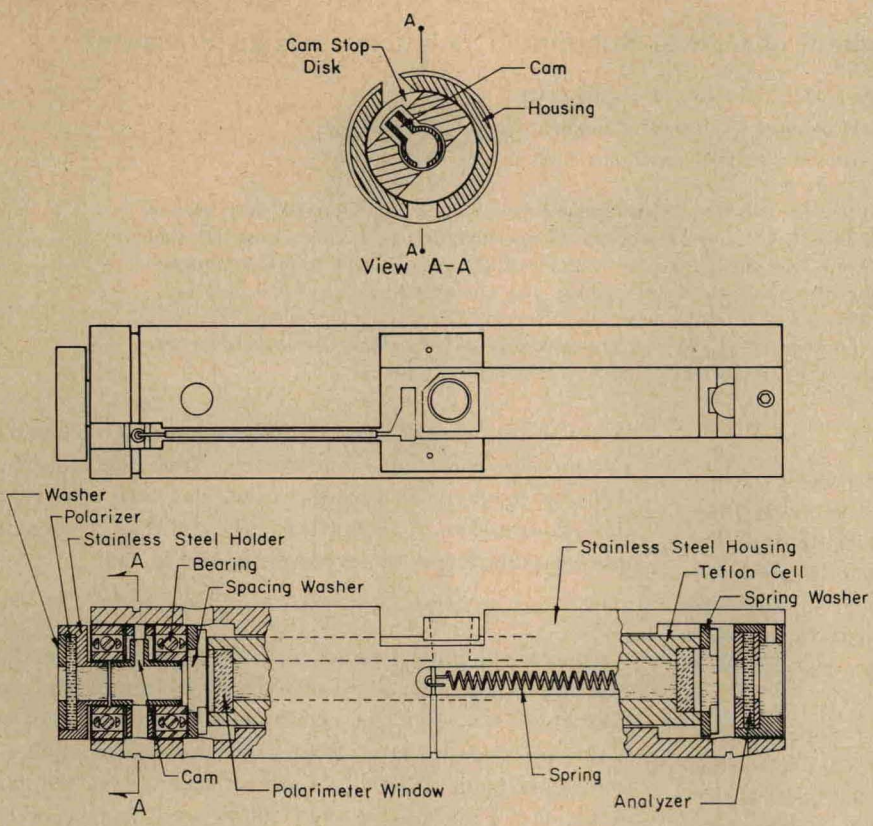

FIG. 1. Polarimeter apparatus designed to fit into a high pressure bomb.[10]

of the polarizer is driven by means of a rod with a bent end. The other end of the rod can be connected to a small cylindrical magnet which extends vertically from the high pressure bomb. The magnet is contained within a drilled, nonmagnetic, stainless steel plug which is surrounded by a solenoid. When the solenoid is not activated, the gravitational force on the mass of the magnet forces the cam to one stop position; and when the solenoid is activated, the vertical movement of the magnet drives the cam to the other stop position.

The cell may be filled with the desired solution using a hypodermic syringe and placed within the high pressure bomb. A small, flexible, bottom-tapered Teflon tube, which serves as a reservoir to accommodate any volume changes, is placed tightly into the aperture of the cell through the vertical opening in the bomb. More solution and, finally, 5 drops of heavy mineral oil, which serves as a separation layer between the solution and the pressure fluid, are added. The entire cell housing is tightly wedged within the bomb so that the lever arm which connects to the magnet rod is situated in the center of the vertical opening. The magnet is then joined to the arm of the rod, and the end plugs are positioned tightly. Pressure fluid (heptane) is pumped into the annular area around the cell, and the vertical plug, which now contains the magnet and holds the solenoid externally, is tightened, filled with fluid, and a small high pressure plug at its top is tightened. The high pressure bomb, now completely assembled, is positioned rigidly between the light source and the photocell of a Beckman DU spectrophotometer.

RESULTS

We have calibrated the instrument at 589 mμ and 20°C using different concentrations of sucrose solutions. The optical path length of the cell was 6.08 cm. A calibration plot of calculated α vs $\tanh(0.576D)$ gave least-squares values of $\beta = 0.074°$ and $\theta_1 = 7.552°$. As expected, the optical rotations α of the various sucrose solutions are found to be insensitive of pressures of 10 000 psi. The increase in con-

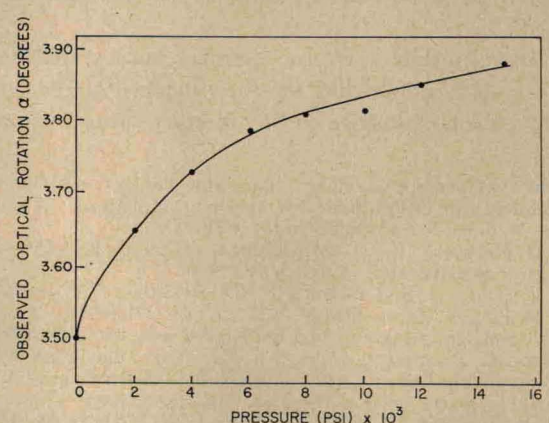

FIG. 2. Effect of increasing pressure on the optical rotation of a 6.8600% (wt./val.) ribonuclease A solution at 589 mμ and 46.8°C. In order to adjust these readings to $[\alpha]$ it is necessary to multiply the ordinate by 23.975.

centration due to compression of the solution with pressure (to 10 000 psi) is of the same magnitude as the sensitivity of the apparatus. Of more interest is the effect of pressure on a solution of ribonuclease A at a temperature in the middle of the region of its thermal transition.[12]

[12] J. G. Foss and J. A. Schellman, J. Phys. Chem. **63**, 2007 (1959).

Here the effect of pressure produces a measurable change in the optical rotation of the solution as shown in Fig. 2.

The buffer solutions used in this experiment was 0.01 M in potassium acid phthalate and 0.15M in KCl. The pH of the ribonuclease solution was adjusted to 2.80 with HCl.