Precise Light-Scattering Studies on Dilute Polymer Solutions. I

Discussion on the Light-Scattering Photometers

Hiroyasu UTIYAMA*, Nagatoshi SUGI**, Michio KURATA** and Mikio TAMURA*

   Various features of the light-scattering photometer are discussed systematically for the purpose of drawing a conclusion what kind of a photometer is most suitable for the precise light-scattering measurements on dilute polymer solutions. Nine light-scattering photometers are selected out of a large number of existing instruments and their characteristics are critically discussed. Some designs which are concluded suitable for our present purpose are illustrated for the projecting optical system, the solution cell, the monitoring optical system, the receiving optical system and the detector electronical system.

INTRODUCTION

   To consider the history of development, the light-scattering method as applied to the physicochemical characterization of macromolecules in dilute solutions was completed like the present form mainly owing to P. Debye[1] and B. H. Zimm until 1950. Relatively speaking, the light-scattering method possessed a considerable advantage at that moment over other methods and the important properties of macromolecules, both synthetic and biological, were clarified in the next ten years by many investigators such as P. Doty[3], for example. In the course of time, however, the remarkable developments in the chemical, biological, and physical methods have been made and we employ the light-scattering method less frequently these days in experiments. But the advantage that only the light-scattering method can provide the information related to the whole spatial distribution of a macromolecule in solution does still hold, which should not be overlooked even if we cannot solely depend on this method.

   The revival of the light-scattering method may be realized if, with use of a more simplified experimental procedure, the experimental error is kept less than one per cent and/or the method can offer more informations of different nature. For example, we have been feeling the pressing need to have a highly precise light-scattering photometer, especially in the study of the excluded volume effect, which can measure the Rayleigh ratio accurately to a very small scattering angle. Because of the difficulty encountered both in the experiment and theory, they depend so much upon each other that the sound progress is obstructed.

   * 内山敬康, 田村幹雄 : Department of Industrial Chemistry, Kyoto University, Sakyo, Kyoto.
  ** 杉 長俊, 倉田道夫 : Laboratory of Polymer Solutions, Institute for Chemical Research, Kyoto University, Uji, Kyoto.

The possibility of the breakthrough of the present situation has recently been shown by G.C. Berry[4], which is naturally to carry out extensive and accurate light-scattering measurements on the well defined samples. We feel quite confident that the physical chemistry of macromolecules will make considerable progress if the accuracy of the light-scattering method is improved.

We initiated the effort to construct a highly precise light-scattering photometer, hopefully to incorporate the laser as the final light source, in 1964. The completion of the apparatus was retarded, however, by various reasons such that the chief investigator was abroad for a couple of years and several problems remain unsolved to incorporate the laser as the light source. But the first stage of the work has recently been completed with success, the detail of which will be published as part II of this series.

In this paper we are going to examine various features of the existing light-scattering photometers to obtain an idea what kind of the photometer can satisfy our present need.

LIGHT-SCATTERING PHOTOMETER

The light-scattering photometers to be discussed in the present paper are listed in Table I. We have chosen them among large number of the apparatus of the similar kind with the criteria that they are constructed for the study of dilute polymer solutions and are unique in some respect. For this reason we ommitted the interesting instruments by J. Piaud[13] for the study of the optical anisotropy,

Table I. The Light-Scattering Photometer discussed in the present paper.

No.	References	Abbreviation
1	B. H. Zimm, *J. Chem. Phys.*, **16**, 1099 (1948)	Zimm[2]
2	B. A. Brice, M. Halwer and R. Speiser, *J. Opt. Soc. Am.*, **40**, 768 (1950)	Brice[5]
3	C. Wippler and G. Scheibling, *J. Chim. Phys.*, **51**, 201 (1954)	SOFICA[6]
4	D. McIntyre and G. C. Doderer, *J. Res. Natl. Bur. Std. U.S.A.*, **62**, 153 (1959)	NBS[7]
5	D. J. Coumou, *J. Colloid Sci.*, **15**, 408 (1960)	Coumou[8]
6	F. J. Baum and F. W. Billmeyer, Jr., *J. Opt. Soc. Am.*, **51**, 452 (1961)	Baum[9]
7	H. G. Jerrad and D. B. Sellen, *Appl. Optics*, **1**, 243 (1962)	Jerrad[10]
8	S. Claesson and J. Ohman, *Arkiv Kemi*, **23**, 69 (1964)	Claesson[11]
9	G. C. Berry, *Mellon Institute Quartely Report*, 1966	Berry[12]

the apparatus for the measurement of scattering of light by thin films[14], and the light-scattering accessory for Beckman DK spectrophotometer[15]. We also limited our survey to the ordinary instruments with the mercury lamp as the light source because the gas laser does not seem yet to refine the apparatus greatly[17]. We are forced to exclude the very attractive apparatus[16] with which S. Katz claimed having carried out the light-scattering measurements on DNA aqueous solution at angles as low as 9 degrees because the detail of the apparatus is not available.

OPTICAL DESIGN

1. Projecting Optical System

The projecting optical system may be classified into three groups. The most popular one (Type A) comprises two achromatic lenses, one (L_1) focusing the image of the mercury arc at the aperture stop and the other (L_2) being located before the cell so that the image of the aperture is focused at the center of the measuring cell* (Fig. 1a). The former condensing lens may be either the cemented doublet (SOFICA, Baum, Jerrad), or the separated doublet[17] (Zimm) in which the neutral filters, the interference filters may be inserted between the two lenses.

mercury arc   lens L_1 aperture stop   lens L_2 diaphragms   cell

(a)

L_1 cylindrical lens

(b)

L_1 L_2 S_2 A_1 L_3 diaphragm S_3
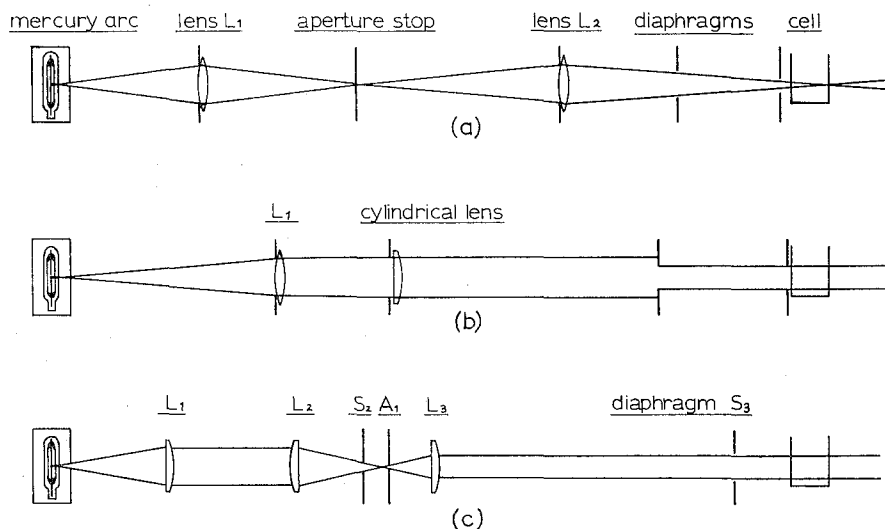
(c)

Fig. 1. Three typical projecting optical systems employed in the light-scattering photometer. (a); Type A (Zimm, SOFICA, Baum, Jerrad), (b); Type B (Brice, Coumou), (c); Type C (NBS, Claesson, Berry).

The uniformity of the brightness and the stability of the incident light beam are realized by employing the proper size of the aperture stop. We may use even two aperture stops separated by couples of millimeters the image of which are both focused at different positions in the cell (Baum).

The primary advantage of this projecting optical system is that the high intensity of the incident beam can easily be obtained without raising the level of the stray light. The slit (the aperture stop) which determine the size of the incident beam in the cell, and the field stop set close to the lens L_2 are both located far from the cell. Hence the stray light level can be kept low by giving attention to use very sharp edges for the aperture stop such as the razor blades, to keep the surface of the lens L_2 scrupulously free from dust or lint, or to set several diaphragms between the lens L_2 and the cell.

* In SOFICA, the image of the aperture stop is focused at the exit face of the measuring cell but we cannot think of the reason exactly.

One apparent disadvantage is the slight divergence of the incident beam, which may cause the overestimate of the optical anisotropy. Usually the divergence amounts to about one degree and the effect, in our opinion, could not be serious because Carr and Zimm[19] obtained fairly accurate depolarization ratio for benzene and carbon tetrachloride*.

The second type of the projecting optical system is represented by the one used in the Brice light-scattering photometer. It contains essentially only one lens at the focal length from the light source. A cylindrical lens added with its axis horizontal reduces the divergens in the vertical direction (Fig. 1b). The size of the incident beam is determined by two rectangular diaphragms 15(V)×15(H), and 15(V)×12(H) which are located between the cylindrical lens and the cell. In this system care is directed mainly to the parallelism of the incident rays at the sacrifice of the intensity. The troublesome stray light from the edges of the diaphragms is a very serious defect, too. The Coumou light-scattering photometer is much better, in this respect, in which another condensing lens is used before the lens as in the type A and the last diaphragm just before the measuring cell has such dimension that the edges do not touch the main light beam. This structure prevents not only producing the stray light from the edges of the last diaphragm but the stray light from the edges of other diaphragms from entering into the measuring chamber.

The last projecting optical system, type C, is used first in NBS and later in the Claesson and Berry light-scattering photometers. The optical design, which was originally constructed by Kushner[20] for the receiver of an absolute light-scattering photometer, was made use of for the projector. The optical system is essentially the same as that of type A except the lens L_3 focuses, at the center of the cell, not the image of the aperture stop (A_4) but of the field stop (S_2) of a definite size located before the aperture stop.

Since the aperture stop is set at the focal point of the lens L_3, the incident beam can be defined distinctly. The most divergent rays are about 0.5° and 0.3° in the vertical and horizontal directions, respectively. We don't think, however, that this telecentric optical system is decisively superior to the type A as long as the calibration of the instruments in various solvents is possible with use of the standard polymer of known molecular weight.

From the above considerations, we conclude that the type A projecting optical system can provide the incident beam of the highest intensity with the lowest level of the stray light provided that the appropriate lenses are chosen to diminish the divergence of the light rays.

To add shortly about the light source, such mercury lamps were used as GE-AH4 (medium pressure, 100 watts) in Zimm and NBS, GE-AH3 (high pressure) in Brice and Baum, MAZDA-ME/D (250 watts) in NBS and Jerrad, Philips-SP500 (high pressure, 500 watts) in SOFICA and Claesson, Philips-103107 (125 watts) in Coumou. Since the information such as the size of the arc, or the life time is not available, further discussion is not given. But it is a matter of course that the lamp of

---

* In the case of Zimm instrument, the divergence of the light rays is three degress in the horizontal direction and five in the vertical direction.

higher and stabler intensity, of smaller size, of longer life time, and of the arc without *"hot-spot-jumping"* is better.

2. The Solution Cell and the Temperature Control

For the measurements of Rayleigh Ratio R_ϑ, the material under study should resume a certain shape and be in thermal contact with liquid to exchange heat for the maintenance of the constant temperature. These conditions are not easy to be satisfied without any interference to the measurement of the scattered light. The material in the liquid state must be confined in a container which is kept in the simultaneous and direct material contact with heat exchanging liquid. Such extraneous material is unavoidably placed in the incident beam which is thereby refracted, reflected, and scattered to the direction different from the incident beam. These stray lights give rise much trouble to the measurements of the scattered light from the material under investigation. Therefore "the design and preparation of a suitable cell to hold the solution under investigation required more care than any other phase of the work" as Zimm pointed out clearly in his paper[2].

There are only very few light-scattering photometers (Zimm, SOFICA, Claesson,* Berry**) which stand the rigorous examination with respect to the above requirements. The measuring cell and the thermostated mantle in the other instruments are not in direct contact except at the bottom of the cell. This structure will demand much effort to bring the solution to the thermal equilibrium and to measure the temperature. Therefore we don't take up such instruments except mentioning that the designs of the cells used in the Jerrad[10] and Baum[9] light-scattering photometers are highly suggestive of the important features that an ideal cell should be embodied with.

The followings are the points we have to keep in mind in designing the light-scattering cell. (1) A small and compact cell easy to handle and simple to construct is preferred. (2) It should be immersed in the liquid the temperature of which is controlled constant with use of the appropriate regulating system and the stirrer. (3) The multiple reflection of the incident or the scattered light fom the interfaces such as glass and air is effectively prevented. (4) Finally the reflection and the scattering of the incident beam, the main source of the stray light, by the material in the path of the incident beam are carefully trapped before entering into the receiving optical system.

The solution cell of Zimm fulfils the above conditions fairly well due to the following arrangements. The cell, a thin-walled (0.2 mm) conical bulb of glass, is immersed in an ordinary 250 ml Pyrex Erlenmyer flask filled with liquid of approximately the same refractive index as the solution in the bulb. The main defect in this cell is the too close location of the surface of the outer cell from the inner solution cell. The light scattered at the front surface of the outer vessel is allowed to enter into the receiving optical system in this structure. Blackened blass shields are introduced in the outer vessel just outside the main beam to block the scat-

---

* Claesson made use of the cell originally designed by Dandliker and Kraut[21], which is referred to later in the text.

** We had to pass Berry's cell without giving further discussion because the detail is not given in the literature.

tering, but the measurements at angles lower than 30° is not possible. The temperature regulation system does not seem extremely useful either, because of its complexity and ineffetive heat exchange.

The light-scattering cell of SOFICA shows the most attractive features in many respects. The cylindrical cell is immersed in a relatively large bath filled with the liquid which has the refractive index close to the glass. The back reflection of the incident light from the exit window in the bath is effectively blocked by introducing a neutral filter in the liquid between the cell and the window. The entrance slit of the receiving optical system is also dipped in the liquid, thereby is the multiple refraction of the scattered light eliminated completely. We feel, however, some further modification will make the experimental procedure much easier.

It is necessary as we can understand from the above discussion that the entrance window in the bath should be located distant from the measuring cell. In the SOFICA cell this condition is indeed fulfilled but the incident beam has now to travel a long path in the outer liquid before entering into the cell. This means painstaking care should be paid not the liquid in the bath to be contaminated with foreign particles all the way through the experiment. By this very reason we hesitate to introduce an motor-driven effective stirrer in the bath for the maintenance of the temperature homogeneity. This we think absolutely necessary.

One compromise to fulfil these incompatible conditions is to incorporate one more removable glass cylinder in the bath with flat bottom and the long entrance tube as employed by Baum and Billmeyer[9] and Dandliker and Kraut[21]. The cell is now positioned at the center of the inner glass cylinder filled with liquid free from dust particles.

The advantage of this structure is three fold. Firstly, we need not pay much care to keep the liquid in the outer bath clean because the incident beam travels only very short path. Therefore the liquid can be agitated strongly with a propeller. Secondly, the stray light from the air-glass interface is completely blocked due to the enough distance to the center of the cell. Finally, since the inner glass cylinder is removable and contains only small amount of liquid, it can be cleaned out of the light-scattering photometer, filled with clean liquid, carried to the temperature equilibrium with the cell set accurately in the position, and finally reset in the light-scattering photometer. Naturally some temperature difference will exist among the cell, the liquid in the inner bath, and that in the outer bath at an elevated temperature, but we need not spend much time to bring the whole system to the temperature equilibrium.

3. The Monitoring Optical System

Since the reduced scattered intensity is defined as a function of the ratio of the intensities of scattered and incident light, we compare the two light levels by a single receiving optical system. This is achieved by rotating the receiving optical system coaxially around the cell, but the accuracy of the measurement is affected seriously by the variation in the incident beam intensity. The instability of the electric power supply and the hot-spot-jumping of the mercury arc are the main reasons of the variation. Therefore we have to either compare the two light

levels simultaneously or incorporate an additional monitoring system to compensate the variation.

Generally speaking, the simultaneous and direct measurement of the intensity of the incident light after it leaves the cell, together with that of the scattered light, is not recommended because the Fresnel reflection from the reference optical system will become a troublesome factor to be corrected for. Therefore we don't employ the interesting null modulation system of Jerrad[10].

The most poplular method is to compensate the fluctuation by comparing the intensities of the scattered light and part of the incident light with use of an appropriate ratio recorder or a potentiometer (see the Electronic Design). Part of the incident beam is reflected by laying, between the aperture stop and the lens L_2 (Fig. 1a), a glass plate with the opening in the center to admit the central part of the beam to pass without interference (Zimm, Coumou). This arrangement will be best provided that the illumination is highly homogeneous.

Strictly speaking, however, the properties of the beam at the center and at the periphery differ as long as the aperture stop has a certain size. We prefer, therefore, to lay a glass plate in the beam although it is somewhat polarized.

Three light-scattering photometers, NBS, Claesson, and Berry, have such type of the reference optical system. Of them, the last is best since the lens in the reference optical system focuses the image of the field stop S_2 (Fig. 1c) at the opal diffuser before the reference photomultiplier tube. Since the image of the field stop S_2 is also focused at the center of the light-scattering cell, this arrangement ensures that the lights received by the reference and receiving optical system are of the same quality.

We may also direct our effort to minimize the variations in the light source intensity. For example, SOFICA incorporates a monitoring photomultiplier which automatically adjusts the high voltage supplied to the measuring photomultiplier to minimize the variation in the photo current. We can not answer, at present, which of the two is better, but the former will be simpler to construct.

4. The Receiving Optical System

As we already stated in the text, we definitely prefer that the front slit of the receiving optical system and the measuring cell are both immersed in the liquid as in SOFICA. We believe that such arrangement relieves us from worrying about the troublesome correction for the reflection, refraction of the incident and scattered light. Needless to say, such effects cannot be suppressed completely because the refractive indices of the solution, glass cell and the liquid in the outer vessel are not exactly the same, but the amounts are far less than those at the air-glass interface. The numerical calibration constant of the apparatus to be multiplied in determining the absolute scattering power may differ depending upon the refractive index of the solution. It may be determined by carrying out the light-scattering measurements with use of the polymeric solute of known molecular weight. By the same token, we don't adhere to the capability of the instrument to measure the absolute Rayleigh factor.

The remaining problem is whether we should employ a lens or lenses in the receiving optical system. Our answer is yes, because "the stray light arising in other parts of the optical system, such as especially the places where the beam

enters and leaves the flask K and reflections of these spots from the walls"[2)], can be effectively blocked. Of the receiver of this kind, Kusher's model[20)] (NBS, Claesson, Berry) and Zimm's model[2)] (Zimm, Coumou) are most widely used. We don't see much difference between the two.

Despite of the advantage of the receiver with a lens or lenses, we think we can do well without, provided the slits, especially the one at the front end, and the diaphragms are made with very sharp edges. Then we can use a front slit of narrow width, which is ultimately necessary to observe the scattered light at very low angles without interfering the incident beam when the distance of the front slit from the cell center cannot exceed a certain size.

In employing the receiving optical system of SOFICA type we have to incorporate additionally a total reflection prism. For the compensation of the change in polarization of the reflected light, two prisms are arranged antisymmetrically. But the second one can be ommitted if the change is corrected for as a part of the polarization correction of the receiving system which includes the correction for the difference in the sensitivity of the photomultiplier tube to the horizontally and vertically polarized light.

ELECTRONICAL DESIGN*

The electronic system frequently in use in the light-scattering photometer can be classified mainly into two groups, one with the reference photomultiplier and the other without it.

In the latter, the electronic system is very simple but the highest precision is not attained because the effect of fluctuation in the intensity of the light source cannot be suppressed. The output currents from the photomultiplier tube are observed either directly on the galvanometer (Brice) or on the recorder after d.c. amplification including the cathode follower circuit (SOFICA). The feedback control of the d.c. power supply for the measuring photomultiplier tube with use of the monitoring photomultiplier to suppress the light source instability (SOFICA) may be recommended. However, there is no reason to adhere to such type if we
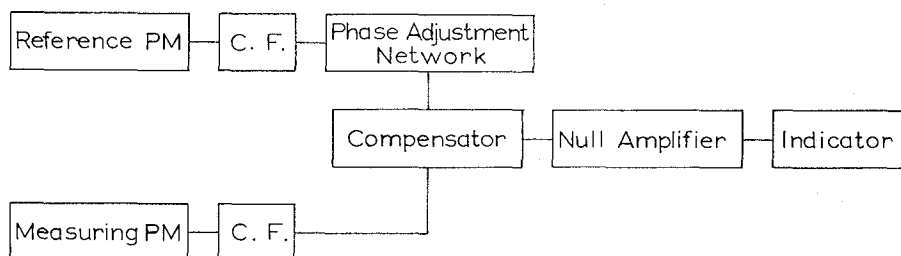

Fig. 2. The block diagram of the a.c. potentiometer in light-scattering photometer.

* Here we limit our consideration to the ordinary electronic system with the vacuum tubes. The recent development of the useful units of the solid-state transistor circuit will make the story completely different. For example, the synchronous rectifier circuit which can be tuned to any desired frequency may be applied. Then, the glass plate to reflect the incident beam into the monitoring optical system will be replaced by a mirror rotating at 10 to 20 c.p.s.[22)]

consider the difficulty in the stable d.c. amplification without increasing the noise level, and the trouble to adjust the zero point of the detector.

In the system equipped with the reference photomultiplier, two outputs from the measuring and reference photomultipliers are compared in the bridge circuit (Zimm, Coumou, Berry) or fed into a ratio recorder after d.c. amplification. Figure 2 represents the block diagram of the a.c. potentiometer. The use of only the modulation of the photocurrents has made it possible to avoid the infavorable situation that the photomultiplier has a large dark current from the amplified thermic emmision of the photocathode. The application of the narrow band null amplifier effectively eliminates the noise of a.c. component[2].

The a.c. signals in the photomultiplier output are produced either by the use of the mercury arc lamp operated on an a.c. power line (120 c.p.s.) (Zimm. Coumou), or by the mechanical modulation of the incident light at 450 c.p.s. while the mercury arc being operated by a d.c. power supply. The latter is naturally preferred for reducing the hum noise level.

The a.c. component of the signals produced at the load register of the reference photomultiplier is fed into one arm of the potentiometer through a cathode follower circuit and the phase adjustment network P. The signals from the measuring photomultiplier are applied to the other arm of the potentiometer. The balance conditioning is determined with a narrow-band null amplifier, which consists of two stage R-C coupled high gain voltage amplifier with a frequency dependent feedback network of the twin-T type. The output is detected either with an electron-ray indicator (Zimm), or with a meter after passing the phase synchronous rectifier.

The null-point method with use of the bridge circuit is best to carry out the precise measurements since the non-linearity of the amplification factor does not affect the results.

From the point of convenience of the experimental procedure, however, it is not of advantage that the direct reading or the recording are not possible. In this respect, the ratio recorder which records the ratio of two input signals by balancing them with a slide resistance and servo-mechanism possibly replaces the potentiometer (Claesson).

The block diagram of the detector electronic system which we conclude most convenient is shown in Fig. 3. The incident beam from the mercury arc operated by the stabilized d.c. power supply is mechanically modulated at 450 c.p.s. The a.c. signal from the measuring photomultiplier is led through an impedance transformer, a narrow-band amplifier and a phase synchronous rectifier, to the ratio recorder.
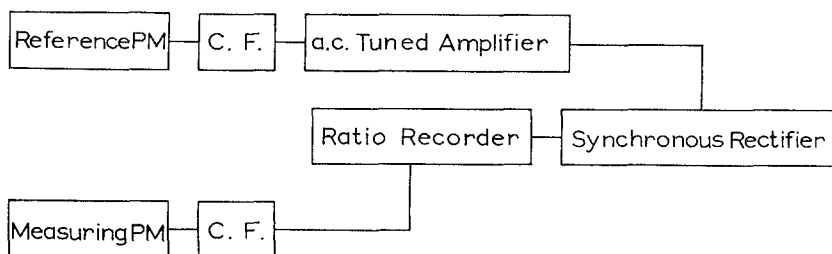
Fig. 3. The block diagram of the detetor electronic system which is recommended in this paper.

H. UTIYAMA, N. SUGI, M. KURATA and M. TAMURA

REFERENCES

(1) P. Debye, *J. Appl. Phys.*, **15**, 338 (1944): *J. Phys. and Colloid Chem.*, **51**, 18 (1947).

(2) B. H. Zimm, *J. Chem. Phys.*, **16**, 1093 (1948): *J. Chem. Phys.*, **16**, 1099 (1948).

(3) P. Doty, "*Biophysical Science-A Study Program*", edited by J. L. Oncley, page 61, John Wiley & Sons, 1959.

(4) G. C. Berry, *J. Chem. Phys.*, **44**, 4550 (1966).

(5) B. A. Brice, M. Halwer and R. Speiser, *J. Opt. Soc. Am.*, **40**, 768 (1950).

(6) C. Wippler and G. Scheibling, *J. Chim. Phys.*, **51**, 201 (1954).

(7) D. McIntyre and G. C. Doderer, *J. Res. Natl. Bur. Std. U.S.A.*, **62**, 153 (1959).

(8) D. J. Coumou, *J. Colloid Sci.*, **15**, 408 (1960).

(9) F. J. Baum and F. W. Billmeyer, Jr., *J. Opt. Soc. Am.*, **51**, 452 (1961).

(10) H. G. Jerrad and D. B. Sellen, *Appl. Optics*, **1**, 243 (1962).

(11) S. Claesson and J. Ohman, *Arkiv Kemi*, **23**, 69 (1964).

(12) G. C. Berry, *Mellon Institute Quarterly Report*, 1966.

(13) J. Piaud, *J. Chim. Phys.*, **59**, 215 (1962).

(14) See, for example, A. Plaza, F. H. Norris and R.S. Stein, *J. Polymer Sci.*, **24**, 455 (1957).

(15) W. H. Beattie and W. S. Gallaway, *Applied Optics*, **4**, 1607 (1965).

(16) S. Katz, *Nature*, **191**, 280 (1961): R. Bjorklund and S. Katz, *J. Am. Chem. Soc.*, **78**, 2122 (1956).

(17) R. C. C. Leite, R. S. Moore, S. P. S. Porto and R. E. Ripper, *Phys. Rev. Letters*, **14**, 7 (1965).

(18) F. A. Jenkins and H. E. White, "*Fundamentals of Optics*", Chapter 9, McGraw-Hill, New York, 1957.

(19) C. I. Carr and B. H. Zimm, *J. Chem. Phys.*, **18**, 1616 (1950).

(20) L. M. Kushner, *J. Opt. Soc. Am.*, **44**, 155 (1954).

(21) W. B. Dandliker and J. Kraut, *J. Am. Chem. Soc.*, **78**, 2380 (1956).

(22) K. Suda, *Private communication*.