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X-Ray Diffraction Studies of the Conformation of Polymer Molecules in Solution.

I. *p*-Iodopolystyrene

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In this paper we present results of an X-ray diffraction study of polymer conformation in solution. A method is described which consists essentially of incorporating atoms of high scattering power into a polymer chain at a preferred position and measuring the interaction of these atoms. The method has been applied to *p*-iodopolystyrene and the results demonstrate directly that a random coil conformation of very short segment length characterizes the structure of the dissolved polymer. Other applications of the method are discussed.

Introduction

The identification of helical conformations¹ in proteins and polypeptides has been well known for some time.^{2a} The persistence of helical structures in solution^{2b} is facilitated for these materials by intramolecular hydrogen bonding and the stereoregularity of component residues. Observations of analogous conformations for synthetic polymers necessarily awaited the preparation of stereoregular materials.³ Moreover, refined analytical techniques⁴⁻⁶ are essential for the detection of helical conformations of at most tenuous stability in solution. The helical form is, of course, not the only one that can be taken up by polymers in solution. Some assume a random coil conformation; others maintain a fairly well-extended linear chain which is not helical.⁷ In many cases, the structural form assumed by the polymer is not well defined, and the experimental evidence cannot be unambiguously interpreted.

As a start in a program of studies on the conformation of polymers in solution, we have carried out a series of measurements on *p*-iodopolystyrene dissolved in chloroform and benzene. The aim of the research was, in essence, to substitute heavy atoms in defined positions along the polymer chain, and by means of X-rays, determine their interaction distance, and in this way characterize the shape of the molecule. This method, an extension of some ideas proposed by Kratky,⁷ will be described in detail later, as will the reasons for choosing this particular polymer as a vehicle for study. To supplement X-ray studies, we also did light scattering and viscosity measurements to elucidate solution properties and infrared absorption studies to detect whether the isotactic polymer in solution possessed the helical structure characteristic of isotactic polystyrene.⁴ It is the purpose of this paper to present our picture of the conformation of *p*-iodopolystyrene in solution and to elaborate the use of the method for studies of isotacticity and other properties of macromolecules.

Theory

The scattering of an assembly of atoms of different kinds is given by⁸

$$I = \sum_m \sum_n f_m f_n \frac{\sin sr_{mn}}{sr_{mn}} \quad (1)$$

(1) We refer to rotational isomers as conformations.

(2) (a) C. Tanford, "Physical Chemistry of Macromolecules," John Wiley and Sons, Inc., New York, N. Y., 1961; (b) P. Doty, A. M. Holtzer, J. H. Bradbury, and E. R. Blout, *J. Am. Chem. Soc.*, **76**, 4493 (1954).

(3) N. G. Gaylord and H. F. Mark, "Linear and Stereoregular Addition Polymers," Interscience Publishers, Inc., New York, N. Y., 1959.

(4) T. Onishi and S. Krimm, *J. Appl. Phys.*, **32**, 2320 (1961), and references.

(5) M. T. Vala, Jr., and S. A. Rice, *J. Chem. Phys.*, **39**, 2348 (1963).

(6) G. Moraglio and F. Danusso, *Polymer*, **4**, 445 (1963).

(7) O. Kratky and G. Porod, *Rec. trav. chim.*, **68**, 1106 (1949), and references.

In this expression I is the intensity, f_m is the structure factor of atom m , r_{mn} is the distance between atom m and n , and $s = (4\pi/\lambda) \sin \theta$, where θ is half the Bragg scattering angle and λ is the wave length. In a solution all varieties of interatomic distances, both inter- and intramolecular, are present and will depend, among other factors, both on the configuration of the solute and solvent molecules themselves, and also on their interaction. In principle a suitable Fourier transformation of eq. 1 will give the distribution of all atoms and distances, but in practice when, as in this case, the solution consists of complex molecules it is difficult to separate the peaks which define each r_{mn} .

To circumvent this difficulty, we adopt the following procedure to characterize the polymer in solution. We first measure the scattering from a monomer solution of the same number of units as the polymer. We next measure the scattering from the polymer solution. When the intensity pattern of the monomer is subtracted, only the pattern arising from the configuration that the polymerized unit has assumed is left, and this eliminates all the scattering that arises from interactions between solute and solvent and from intramolecular preferred distances that would be present in both solutions. Since the X-ray absorption will be the same in both cases, it is permissible to subtract intensities in this way. Thus, if the polymer in solution retains a rigid conformation for large distances along the chain, by substituting heavy scattering atoms such as iodine into monomer residues in the polymer, the subtraction process will leave as contributors to eq. 1 only those distances r_{mn} characteristic of the row of substituted atoms in the chain.

Two cases come to mind at once for the application of this method. The first is to examine the random coil conformation. If the randomness is characterized by a short Kuhn segment length,⁹ then we should expect little difference in scattering between monomer and polymer solution. However, if the polymer shape is such that long regular segments are present, then a definite I-I interaction would show up in the differential intensity curve and also in the radial distribution function (RDF) derived therefrom. For example, if we envisage an isotactic polymer which is fully extended in solution we expect that the *differential* intensity would be given by

$$I = Nf^2 \left[1 + \int_0^\infty 4\pi r^2 (\rho(r) - \rho_0) \frac{\sin sr}{sr} dr \right] \quad (2)$$

where $\rho(r)$ is the density of iodine atoms at a distance r .

(8) N. S. Gingrich, *Rev. Mod. Phys.*, **15**, No. 1, 90 (1943).

(9) W. Kuhn and H. Kuhn, *Helv. Chim. Acta*, **26**, 1394 (1943).

and ρ_0 is the average density of these atoms, considered to be completely random. The Nf^2 term refers now to the number N of iodine atoms of scattering factor f . By the usual Fourier transformation we get

$$4\pi r^2 \rho(r) = 4\pi r^2 \rho_0 + \frac{2r}{\pi} \int_0^\infty \left(\frac{I}{Nf^2} - 1 \right) s \sin rs ds \quad (3)$$

which gives the spatial distribution of iodine atoms along the chain. In the case of *p*-iodopolystyrene there would be a contribution from the scattering between carbon atoms in adjacent benzene rings but since the scattering power of the iodine atoms is so much greater this contribution would be trivial. For a completely extended isotactic polymer eq. 3 predicts a value of 2 for the number of nearest neighbors ($4\pi r^2 \rho$) at a distance r equivalent to the I-I isotactic distance, while for a similarly extended atactic polymer the number would be less than this, and in addition there should be a characteristic I-I distance for the atactic material. Other cases, such as the helix, would also be expected to exhibit characteristic scattering and our labeling technique should lead to definite information about these structures as they exist in solution.

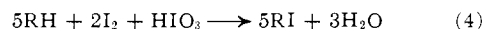
Experimental

The diffraction patterns were measured with a North American Philips diffractometer equipped with a liquid diffraction cell, as previously described.¹⁰ However, in the earlier work on aqueous solutions, the solvent, water, did not diffuse into the thin polymer film bounding the upper surface of the solution; and thus we did not encounter the difficulties which soon became apparent when organic solvents were used. Many of the films, notably polystyrene, were soluble in benzene and chloroform. Mylar film gave a broad intense peak in the angular region centered around $2\theta = 12^\circ$ (Mo $K\alpha$) radiation, where we were particularly interested in getting accurate measurements of the scattering. We finally resorted to 1-mil polyethylene film in which, although they did interfere somewhat with the scattering, the reflections were quite sharp, and could easily be corrected for. However, we were still confronted with the effect of penetration of the membrane by organic solvents. In the Brentano *para*-focusing geometry used it is important that the scattering surface be flat and exactly at the center of the scattering circle. The absorption of solvent by the membrane caused it to swell when the cell was filled and changes in intensity up to 25% resulting from the displacement of the surface were not uncommon. To get around this source of error, we irradiated the polyethylene in a Van de Graaff generator to stabilize the film by cross linking it, thus decreasing the swelling. It was found that a dose of 100 Mrads accomplished this; further irradiation made the films too brittle. The final procedure adopted, which gave very reproducible results, was to stretch the irradiated film over the top of the cell and keep solvent in the cell at all times, even between experiments. If we did not do this, the films would sag when allowed to dry and could not be used again.

The diffraction patterns were taken with Cu $K\alpha$ radiation for the lower angles, and Mo $K\alpha$ radiation for the larger angles. To ensure that no change in intensity due to evaporation of the solvent took place, checkpoints were chosen at various angles on the diffraction pattern, and after every fifth reading was taken, a check was made at one of these points. If any change greater than 1% were noticed, the solution was changed and the readings repeated.

Iodine labeled polystyrenes were prepared by the iodination of atactic and isotactic polystyrene (The Borden Chemical Co.) according to the procedure of Braun.¹¹ The polymer was dissolved in nitrobenzene and heated to 90° for 30 hr. with iodine, iodic acid, and sulfuric acid catalyst. The isotactic polystyrene was initially insoluble in the reactant mixture but was progressively solubilized on iodination. This process, in which iodine

substitutes in the *para* position of pendant benzene rings, is described by the equation



The extent of iodination was ascertained by gravimetric analysis (Carius bomb technique¹²). In addition, direct measurements of iodine content were effected by analysis of infrared spectra of iodinated polystyrenes.^{11,13a} Thus, iodine content was found to correlate well with the ratio of infrared absorption bands at 820 and 758 cm^{-1} . Nuclear magnetic resonance spectra (left-hand side of Fig. 4) indicated *para* substitution.^{13b} For the atactic polystyrene it was found that complete *para* iodination was effected by using a 30% stoichiometric excess of iodine and iodic acid. Analogous treatment of the isotactic polymer yielded less efficient iodine substitution (probably because the isotactic polystyrene is initially insoluble in nitrobenzene) unless the isotactic material was first degraded slightly by ultrasonic treatment in nitrobenzene suspension. Retention of tacticity by the resultant isotactic *p*-iodopolystyrene was confirmed by n.m.r. measurements,^{13b} and will be subsequently discussed. The X-ray studies reported involve almost completely iodinated atactic and isotactic polystyrenes. The concentrations used were those equivalent to 0.734 g.-atom of iodine per liter.

Molecular weights, radii of gyration, and second virial coefficients were determined from light scattering measurements on dilute solutions of isotactic and atactic iodinated polystyrenes in benzene using a Sofica photometer.^{14,15} In addition, refractive increments were measured on the same solutions with a Water's differential refractometer, and intrinsic viscosities in Cannon-Ubbelohde semimicro dilution viscometers. Results are illustrated in Table I and will be subsequently elaborated.¹⁶ Essentially, we confirm previously observed decreases in intrinsic viscosity at constant degree of polymerization.¹¹ That is, there is no chain degradation associated with increasing degrees of iodination, but polymer-solvent interactions are minimized.

TABLE I

SOLUTION PROPERTIES OF IODINATED POLYSTYRENES				
%I ^a	\bar{M}_w^b	\bar{p}^b	B^b	$(\eta)^c$
Atactic	236,000	2.44×10^{-6}	7.0×10^{-4}	0.84
42.6	369,000	2.40	3.6	.39
51.5	462,000	1.65	2.1	.25
Isotactic				
50.8	1.1×10^6	2.89	3.2	.30

^a Weight of iodine corresponding to theoretical monosubstitution = 55.2%. ^b Light scattering: \bar{M}_w = weight average molecular weight; \bar{p} = radius of gyration in cm.; B = second virial coefficient. ^c (η) = intrinsic viscosity in dl./g.

Results and Discussion

Figure 1 shows the diffraction patterns of atactic *p*-iodopolystyrene and of *p*-iodotoluene at the same concentration in benzene. The two patterns are indistinguishable. An equivalent result was obtained in chloroform solutions. This indicates directly that there is no arrangement of iodine atoms of sufficient regularity to give rise to a scattering different from that in a purely random array of *p*-iodostyrene groups. In Fig. 2 we show the scattering of three solutions, taken with Mo $K\alpha$ radiation. Curve 1 is the pattern of toluene in chloroform, curve 2 that of toluene itself, and curve 3 that of chloroform. Curves 1 and 3 have been scaled down to a common intensity at $2\theta = 20^\circ$, and curves 1 and 2 at $2\theta = 5^\circ$. The absorption in these three solutions is not comparable, and therefore the intensities are only relative. The peak at $2\theta \sim 9^\circ$ in curves 1 and 2 is characteristic of intramolecular scattering of the benzene rings, and is absent in curve 3.

(12) Dr. C. Tiedcke, Teaneck, N. J.

(13) (a) J. P. Luongo, personal communication; (b) F. A. Bovey, personal communication.

(14) K. A. Stacy, "Light Scattering in Physical Chemistry," Academic Press, New York, N. Y., 1956.

(15) C. Wippler and G. Sheibling, *J. chim. phys.*, **51**, 201 (1954).

(16) R. Salovey and M. Y. Hellman, paper in preparation.

(10) G. W. Brady, *J. Chem. Phys.*, **27**, 304 (1957); **28**, 1371 (1958).

(11) D. Braun, *Makromol. Chem.*, **30**, 85 (1959).

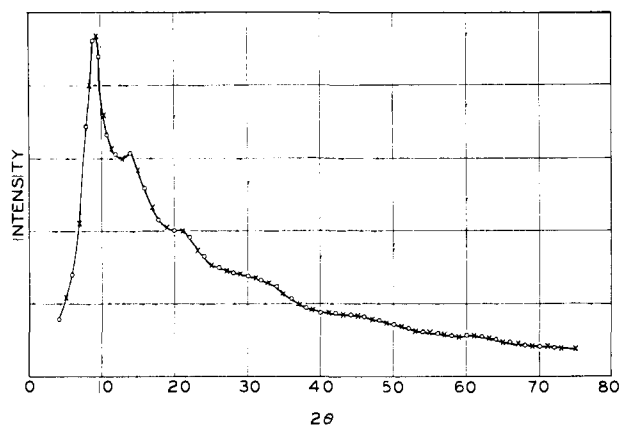


Fig. 1.—Diffraction patterns of atactic *p*-iodopolystyrene in benzene (X) and an equivalent concentration of *p*-iodotoluene in benzene (O).

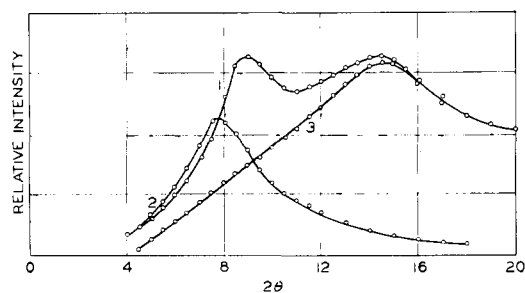


Fig. 2.—Curve 1 is the diffraction pattern of toluene in chloroform, curve 2 that of toluene itself, and curve 3 that of chloroform. The toluene concentration in curve 1 is one-half that of the equivalent curve of *p*-iodotoluene in Fig. 1 and 3.

These results are presented to show that since the ring scattering can be detected the fact of the coincidence of the two higher curves in Fig. 1 is not an artifact because the much higher scattering power of the iodine atoms should make any I-I interaction difference between monomer and polymer result in a considerable difference in the intensity pattern.

It may be surmised (incorrectly) that we are unable to detect any preferred scattering in Fig. 1 because the steric arrangement of iodobenzene groups is essentially random in an atactic configuration. However, it is evident that this is not the final explanation because there should be enough I-I interactions, even in this configuration, to give rise to additional peaks in the diffraction pattern of the polymer. To anticipate our further results, we will find that it is not tacticity but rather the conformation of the backbone itself which is responsible for the absence of characteristic interactions in the polymer solution.

Figure 3 shows the intensity patterns of isotactic *p*-iodopolystyrene and the equivalent concentration of *p*-iodotoluene in chloroform (the isotactic polymer is insoluble in benzene). Here, where the regularity of spacing of the iodine atoms along the chain should be maximal we find again that there is no detectable difference in the intensity patterns of the monomer and polymer solutions. What this means in terms of eq. 3 is that the $[\rho(r) - \rho_0]$ term in the integral is the same for both solutions. Diffraction patterns measured at concentrations of one-half and one-quarter of those of Fig. 1 and 3 showed the same match between monomer and polymer, indicating that there was no concentration

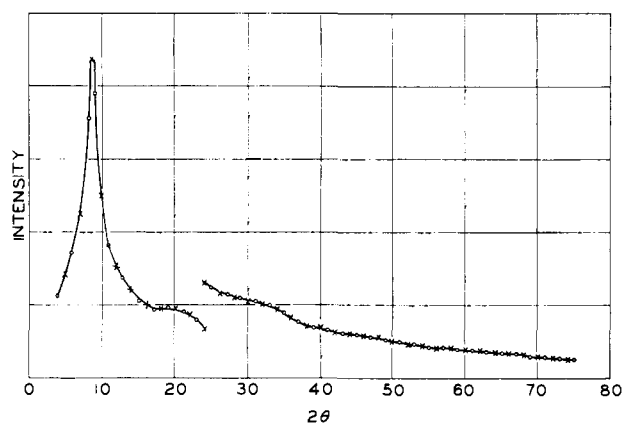


Fig. 3.—Diffraction patterns of isotactic *p*-iodopolystyrene in chloroform (X) and an equivalent concentration of *p*-iodotoluene in chloroform (O). The ordinate side has been increased at $2\theta = 25^\circ$ to improve the detail at the larger angles.

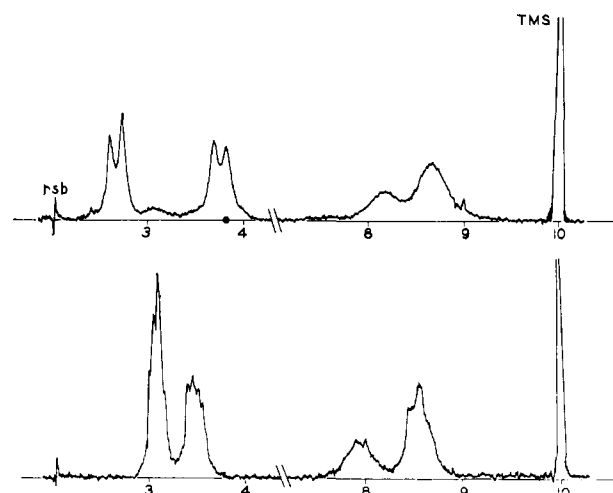


Fig. 4.—N.m.r. spectra. Top, iodinated isotactic polystyrene in carbon disulfide. Bottom, isotactic polystyrene in carbon tetrachloride. Scale is in τ -units (parts per million measured downfield from tetramethylsilane [TMS], the τ of TMS being arbitrarily assigned a value of 10.00); rsb refers to a reference side band.

dependence of the conformation of the polymer chain that would cause it to change on dilution.

The identity of X-ray intensity patterns for the stereoregular and atactic *p*-iodopolystyrenes upon dissolution cannot be attributed to any loss of tacticity by the former. Regardless of iodination, the isotactic and atactic polymers are differentiated by their n.m.r. spectra.^{13b} The right-hand side of Fig. 4 shows the separation of CH and CH₂ resonances characteristic of isotactic polystyrene and, presumably, isotactic *p*-iodopolystyrene as well. There is no separation for the corresponding atactic materials.

From an analysis of solid state and solution infrared spectra of polystyrenes, before and after iodination, we may infer some information concerning the conformation of the polymer chain in solution.^{13a} A characteristic splitting of the band at 1065 cm.⁻¹ has been identified with helix formation.⁴ This band is indicated in Fig. 5 for atactic and isotactic polystyrenes in the solid state and in carbon disulfide solutions, before and after iodination. It is to be noted that the splitting is present only for the isotactic polystyrene, and that it has disappeared in the iodinated polymer. Thus, to sum-

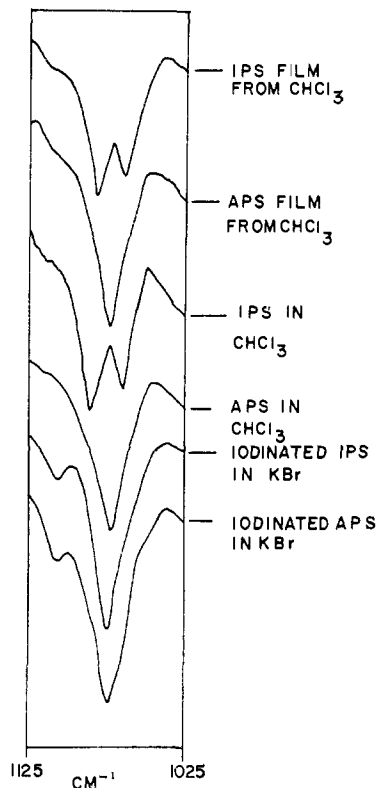


Fig. 5.—Infrared splitting of the 1065-cm.⁻¹ band of isotactic polystyrene (IPS) and atactic polystyrene (APS). Note: iodinated polystyrenes are identical in potassium bromide and in chloroform solution.

marize, n.m.r. results indicate that, on iodination, the local configuration remains isotactic. However, infrared spectra suggest a disappearance of the helical character of the chain.

The X-ray results are consistent with these observations and show further that the arrangement of substituted iodine atoms along the chain is essentially equivalent to that of the monomer solution. This indicates, first, that the polymer conformation is that of a random coil, and second, that the statistical segment length characterizing the coil must be small because even for a random conformation, if long regular arrays of iodine atoms were present within the segments, it is highly probable that they would be detected. We next proceed to a calculation of the statistical segment length and we will discuss this in the terms of Kratky and Porod,⁷ who define the persistence length of a segment as being equal to half a Kuhn element. The Kuhn element is a hypothetical statistical unit which is completely random in orientation within the polymer chain. Although the polymer may not be treated as a freely jointed array of monomer residues because of valence angle restrictions and hindered rotation about single bonds, these restrictions do not preclude random flight statistics⁹ and the actual polymer chain may be described by an equivalent freely jointed chain composed of uncorrelated statistical segments. Following the notation of Flory,¹⁷ a real polymer chain composed of n bonds, each of length l , may be replaced by a random array of n' Kuhn elements of length l' so that

$$nl = n'l' = L$$

The contour length L of a chain is the total length of all bonds laid end to end.² The root mean square (r.m.s.) end-to-end distance of the real chain is given by

$$\sqrt{\bar{r}^2} = l'(n')^{1/2}$$

It follows that $\bar{r}^2/L = l'$

We note that if the main chain were freely jointed $L/\sqrt{\bar{r}^2}$ would equal the square root of the degree of polymerization ($n^{1/2}$), and l' would equal the bond length l . The quantity $L/\sqrt{\bar{r}^2}$ has been designated the degree of coiling.⁹

From the light scattering measurements in dilute solution (Table I) we can evaluate the quantities $\sqrt{\bar{r}^2}$ and L for the isotactic polymer. Thus, for a weight-average molecular weight of 1.1 million and a z average radius of gyration of 289 Å, we derive a z average contour length of 24,300 Å, and an r.m.s. end-to-end distance of 710 Å.¹⁴ This indicates a Kuhn length of about 20 Å, and therefore a persistence length of 10 Å. It is interesting to note that an analogous calculation for atactic polystyrene of comparable molecular weight in benzene gave a value of 34 Å, for l' . In the latter case the chain is extended because of interaction with the solvent. When thermodynamic interactions between polymer segments and solvent exactly compensate the influence of volume exclusion (so-called θ -solvent conditions), the chain molecule is referred to as unperturbed and assumes a random flight conformation. The low solubility of *p*-iodopolystyrenes in benzene and chloroform suggests that unperturbed dimensions probably obtain. Moreover, the decrease in intrinsic viscosity, radius of gyration, and second virial coefficient on iodination (Table I) indicate that polymer solvent interactions, which tend to expand the chain, are decreased and that θ -conditions are approached.

Since in the helical form¹⁸ the I-I distance is 6.7 Å, it becomes even clearer why the intensity pattern of the polymer solution is similar to that of the monomer. Since the persistence length measures the distance along the chain at which complete randomness results, it can be seen from the calculation that this is of the same order of magnitude as the I-I distance, and therefore we would not expect to see any difference in scattering. The I-I distance would probably be different in the nonhelical form, but it would almost certainly not change enough and assume enough regularity to offset the effect of the highly random conformation of the polymer chain. Indeed, the calculated coherence length is so small that it is questionable whether the Kuhn statistics even apply because it is assumed in applying this model that a broad distribution of segment lengths is appropriate.⁷ Since the preferred length approaches that of the individual monomer residues in the chain, the idea of a segment length spectrum wide enough to give tangible reality to such a distribution becomes tenuous. However, the essential fact to be noted is that the X-ray results confirm the statistical calculations in that both lead to a very short persistence length. Thus, the X-ray method proves to be a powerful and direct measurement of the degree of regularity of polymers in solution.

The next step in our research will be to apply this method to other polymers which would be expected to

(17) P. J. Flory, "Principles of Polymer Chemistry," Cornell University Press, Ithaca, N. Y., 1953.

(18) G. Natta, P. Corradini, and I. W. Bassi, *Nuovo Cimento, Suppl.*, **14**, 1, 69 (1966).

exhibit less randomness in solution than the *p*-iodopolystyrene-solvent system. In our next communication we will describe our results on 3,5-dibromopoltyrosine, a synthetic polypeptide, which optical rotation studies¹⁹ indicate has an extended helical form in solution.

Acknowledgments.—G. W. B. wishes to acknowledge the assistance of Professor Herbert Morawetz,

(19) J. Applequist, personal communication.

who in the course of a stimulating discussion pointed out the possibilities of investigating conformation, configuration, and isotacticity by means of X-ray measurements on polymer solutions. We are indebted to F. A. Bovey and F. P. Hood for n.m.r. measurements, to J. P. Luongo for infrared spectra, and to Mrs. M. Y. Hellman for light scattering and viscosity determinations. The work benefited greatly from the interest shown during the course of the research by F. A. Bovey.

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Light Scattering and the Structure of Pure Water

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The results of light scattering by pure water should have a bearing upon the acceptability of the various proposed structures of liquid water. Despite the difficulty of experimental determination it seems that the excess turbidity, due to structural heterogeneities, above that due to pressure fluctuations is very small. This does not seem compatible with a structure involving compact "icebergs" separated by a "liquid" nor with a large volume fraction of molecule-sized random vacancies. A network with predominantly filled cavities seems more compatible with the experiments.

The unique properties of liquid water are well known and have long been attributed to the association of its molecules. At present there seems to be widespread agreement with Frank's¹ view that this association leads to "flickering" structures of many water molecules. However, the nature of these structures is still uncertain. They may be viewed as compact iceberg-like clusters² on the one hand, or, on the other, as honeycomb networks of cages.³⁻⁶ In either case unassociated water molecules constitute at any instant an important fraction of the total, either separating the icebergs or located inside and presumably also between⁵ the cages. The cage structures may be either essentially filled with unbonded molecules³ or be vacant to a significant extent.⁴⁻⁶

Both approaches have been reasonably successful in accounting for the adduced experimental evidence, particularly the thermodynamic properties of the liquid and its X-ray scattering which can be interpreted in terms of the average radial neighbor distribution as shown by Debye in 1930.⁷ Hence preference for either of these models is based largely on structural grounds and analogies. The purpose of the present paper is to bring into this discussion the experimental argument of light scattering which does not seem to have been used heretofore, and to show that it favors the network model without many vacancies over the compact iceberg one.

The turbidity of pure water is due to its inhomogeneities in refractive index which in turn stem from two independent sources which give separate additive contributions to turbidity. One of these contributions is made by fluctuations in density caused by pressure fluctuations due to thermal agitation, the other is

made by local differences in structure which occur independently of pressure variations. As established by Einstein,⁸ the contribution of the former to the turbidity can be calculated from the macroscopic compressibility and the macroscopic variation of refractive index with density. Whereas these values in turn depend on structure, so that this part of the turbidity could in principle be calculated from models, we shall not pursue this aspect. It has been discussed for example by Némethy and Scheraga² with respect to compressibility. By using the experimental quantities we take into account automatically the effect of fluctuations in pressure upon structure.

Thus the turbidity due to fluctuations in pressure calculated from the experimental quantities can be considered as a base line and any excess above it can be assigned to structural heterogeneities or experimental error. The excess turbidity to be expected from structure will depend on the size of the postulated heterogeneities, on their refractive index contribution, on their concentration, and on the randomness of their spacing as they occur at constant pressure.

As shown in more detail below the turbidity due to pressure fluctuations is about 1.56×10^{-5} cm.⁻¹ and the measured value is about 1.76×10^{-5} leaving only a very small margin of about 0.2×10^{-5} for any excess turbidity due to structure.

Compact "icebergs" having an ice structure and comprising a score or more of water molecules represent relatively large heterogeneities with a significant refractive index contribution and in the absence of any long range forces should be spaced randomly. One can expect therefore that they would contribute a significant excess turbidity. In fact as shown below for a specific model, this excess turbidity, though still small, is of the same order as the total experimental turbidity of water and several times larger than the experimental excess turbidity evaluated above.

In the network model the heterogeneities are much smaller, of the order of a molecule instead of an iceberg,

(1) H. S. Frank and W. Y. Wen, *Discussions Faraday Soc.*, **24**, 133 (1957);

H. S. Frank, *Proc. Roy. Soc. (London)*, **A247**, 481 (1958).

(2) G. Némethy and H. A. Scheraga, *J. Chem. Phys.*, **36**, 3382 (1962).

(3) L. Pauling, "The Nature of the Chemical Bond," 3rd Ed., Cornell University Press, Ithaca, N. Y., 1960, p. 472.

(4) M. D. Danford and H. A. Levy, *J. Am. Chem. Soc.*, **84**, 3965 (1962).

(5) H. S. Frank and A. S. Quist, *J. Chem. Phys.*, **34**, 604 (1961).

(6) R. P. Marchi and H. Eyring, *J. Phys. Chem.*, **68**, 221 (1964).

(7) P. Debye and H. Menke, *Physik. Z.*, **31**, 797 (1930).

(8) A. Einstein, *Ann. Physik*, **33**, 1275 (1910).