

FLUORESCENCE STUDIES OF CONFORMATIONAL MOBILITY AND OF THE MUTUAL INTERPENETRATION OF FLEXIBLE CHAIN MOLECULES

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Abstract- Intramolecular excimer fluorescence may be used to estimate rates of conformational transitions with low activation energies. Fluorescence spectra can, therefore, be employed to show that the conformational mobility in the backbone of polymers in dilute solution is similar to that of a low molecular weight analog. The same technique yields data on the dependence of conformational mobility on molecular structure and the viscosity of the medium. Nonradiative energy transfer between two macromolecules carrying appropriate fluorescent labels can be interpreted in terms of the mutual interpenetration of the polymer chains. This technique was applied to the study of polymer solutions and to the characterization of the compatibility of polymers in bulk.

INTRODUCTION

Fluorimetry, which has long been recognized as a powerful tool for the study of biological macromolecules (1) has also found a wide variety of applications in investigations of synthetic polymers (2-4). Such studies may utilize (a) fluorescence quenching, (b) depolarization of fluorescence, (c) excimer fluorescence and (d) nonradiative energy transfer between two chromophores. As we shall see, excimer fluorescence is particularly valuable in the study of certain problems concerning the rate of hindered rotation in the backbone of chain molecules, while intermolecular nonradiative energy transfer between chromophore labels attached to polymer chains can be interpreted as reflecting their mutual interpenetration.

MECHANISM OF CONFORMATIONAL TRANSITIONS OF CHAIN MOLECULES IN DILUTE SOLUTION

Hindered rotation in the backbone of a chain molecule presents a conceptual difficulty as represented schematically in Fig. 1. If the process involves

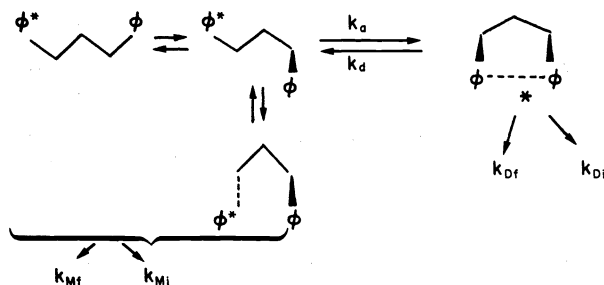


Fig. 1. Schematic representation of conformational transitions in polymer chains. (a) A single rotation around a bond in the chain backbone. (b) Two correlated rotations in a crankshaft-like motion.

rotation around one single bond, then a large part of the chain has to move through a viscous medium resulting in a prohibitively large dissipation of energy. It was, therefore, suggested that two conformational transitions are correlated in time, so that only a short segment of the chain has to move in a "crankshaft-like motion". This concept was originally advanced to interpret the behavior of polymers in bulk (5,6), but it was later also employed in theoretical considerations of the conformational mobility of flexible chains in dilute solutions (6-10). However, here another difficulty is encountered. Simultaneous rotation around two bonds should require an activation energy twice as large as for the hindered rotation around a single bond in an analogous small molecule (6,10); yet, data obtained by depolarization of fluorescence (11,12) and by dielectric dispersion (13,14) yielded generally activation energies of 3 to 5 kcal/mole, quite similar to those for liquid butane (15) or 2,4-diphenylpentane (16), the low molecular weight analog of polystyrene. (Only in a single study of polystyrene solutions by sound absorption was a value as high as 7.5 kcal/mole estimated for the activation energy of hindered rotation (17), but this technique led to strikingly different results in another laboratory (18)). We are facing here a disturbing discrepancy between the prediction of theory and the experimental data.

Clearly, it is most desirable to obtain comparative data on the rate of hindered rotation in the backbone of a polymer and in a low molecular weight analog. This cannot be accomplished by either fluorescence depolarization or dielectric dispersion measurements, since these effects reflect conformational transitions only in macromolecules so large that rotational diffusion of the molecule may be neglected; in small species the effect of rotational diffusion will dominate any effect due to conformational change. Other techniques which have been used for the study of the conformational mobility of polymers, i.e., NMR relaxation and motional narrowing of the ESR spectrum of spin labels attached to the macromolecular chains, suffer from the same limitation.

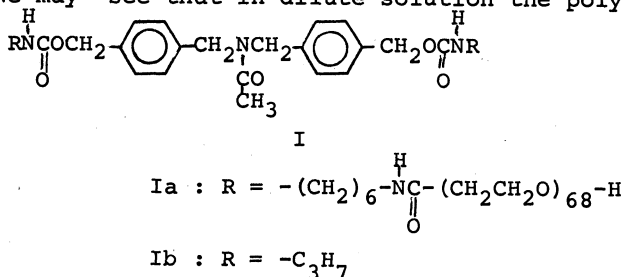
Intramolecular excimer fluorescence offers a convenient tool for solving this problem. This phenomenon was first observed for 1,3-diphenylpropane, which exhibits in addition to the normal phenyl fluorescence peaking at 285 nm an additional emission band with a maximum at 335 nm, which may be shown to be due to the exothermic formation of a sandwich complex between an excited phenyl group with a phenyl group in the ground state (19). Since the conformation required for excimer formation would have a prohibitive energy requirement if both phenyl groups were in their ground state, we have to assume that the excimer forms by a conformational transition after excitation of one of the phenyls. This interpretation is supported by the observation that excimer formation in 1,3-diarylpropanes is eliminated in viscous solvent media (20). We may then represent the various processes by Scheme I, where k_{Mf} and k_{Mi} are rate constants for the fluorescence and the



Scheme I

nonradiative deactivation of the "monomer", k_{Df} and k_{Di} are the corresponding quantities for the excimer, while k_a and k_d are rate constants for excimer formation and dissociation. At relatively low temperatures $k_d \ll k_a$. We may also assume that excimer formation does not perturb appreciably the equilibrium between the various monomer conformations, since the monomer emission in 1,3-di(2-naphthyl)propane was found to exhibit simple exponential decay (21). The relative emission intensity of the "monomer" and excimer, I_M/I_D , should then be proportional to k_{Mf}/k_a' where $k_a' = \alpha k_a$ and α is the fraction of the monomer which exists at equilibrium in the tg conformation from which the excimer can form by a single hindered rotation (20). Note that I_M/I_D will be independent of any motion of the molecule as a whole, so that the fluorescence spectrum can be used to compare k_a in a polymer which contains in its backbone a group capable of forming an intramolecular

excimer with the k_a value observed in an analogous small molecule. A comparison of I_M/I_D of the polymer Ia and its analog Ib is shown in Fig. 2(22). We may see that in dilute solution the polymer and its analog



behave almost identically, i.e., the rate of the conformational transition

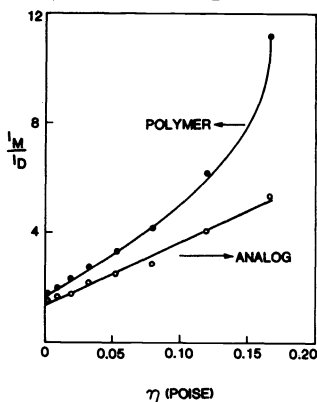


Fig. 2. I_M/I_D ratio of polymer Ia and analog Ib at 25°C in methylene chloride and in methylene chloride solutions of poly(ethylene oxide) with $\bar{M}=3000$. The most viscous solution contained 70 weight-% PEO.

required for the excimer formation is not appreciably affected by incorporation of the dibenzylacetamide into the backbone of a polymer chain. However, the conformational mobility of the label is reduced much more rapidly by an increasing polymer concentration when it is incorporated into the polymer chain than when it is part of a small molecule. This result confirms conclusions arrived at in a previous study in which the quantum yield of the photochemical isomerization of azobenzene residues incorporated into the backbone of polymer chains were compared with the same process in small molecules (23). I believe that these data definitely disprove the mechanism of crankshaft-like motions for the conformational transition of flexible chain molecules in dilute solution. It may be noted that a recent computer simulation of conformational transitions in chain molecules led also to the conclusion that the activation energy is no higher than a single barrier height (24). It remains to be seen how a theory of the conformational transitions of chain molecules in dilute solutions can be devised which is consistent with these results.

THE DEPENDENCE OF THE RATE OF CONFORMATIONAL TRANSITIONS ON THE VISCOSITY OF THE MEDIUM

The dependence of conformational mobility on the solvent medium is of interest to the polymer chemist because of its intimate relation to the concept of an "internal viscosity" of chain molecules. This concept was first introduced by Kuhn and Kuhn (25) who pointed out that if the ends of a coiled chain are to be separated at any given rate in response to an applied force, the resistance will have a friction-like component due to the need to surmount potential energy barriers associated with the conformational transitions. Kuhn and Kuhn believed that this "internal viscosity" is an intrinsic property of the chain, independent of the solvent, but data on the viscoelasticity of dilute solutions (26,27) make it clear that in viscous solvents the internal viscosity of polymer chains tends to be proportional to the viscosity of the medium. This is not surprising since any conformational transition requires necessarily a displacement of solvent molecules. Peterlin suggested (28) that the internal viscosity should be of the form $A+B\eta_0$ where A is the contribution due to potential barriers in

conformational transitions, η_0 is the viscosity of the solvent and the coefficient B depends on the bulk of the groups which have to move through the solvent during a conformational transition. Similar conclusions were later arrived at by Cerf (29) and McInnes (30). It would be expected that the internal viscosity of polymeric chains, involving hindered rotations in response to a mechanical force, should have a similar dependence on the viscosity of the medium as the rate of spontaneous conformational transitions due to thermal motions. In fact, data on the depolarization of fluorescence of a polystyrene labeled with a suitable chromophore in media of varying viscosity led to the conclusion that the rate of conformational change is inversely proportional to $A+B\eta_0$ (12). Now, with the availability of excimer fluorescence as an experimental tool, the dependence of conformational mobility on viscosity and molecular structure can be much more precisely defined using suitably chosen model compounds. Fig. 3

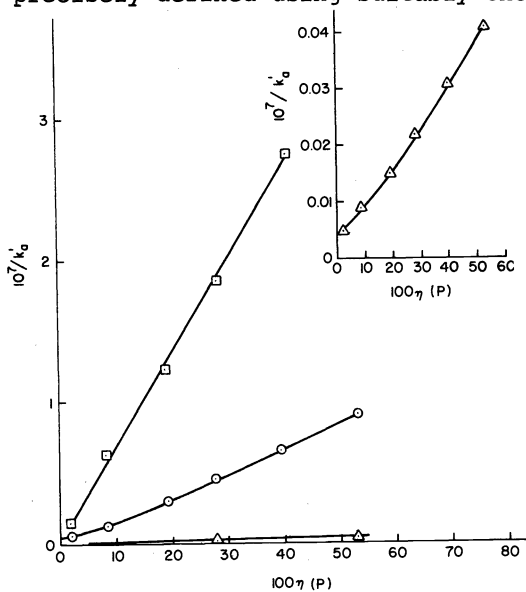


Fig. 3. Viscosity dependence of K'_a in compounds of the type $\text{ArCH}_2\text{OCH}_2\text{Ar}$ with $\text{Ar}=\text{phenyl}(\Delta)$, $\text{Ar}=\text{1-naphthyl}(O)$ and $\text{Ar}=\text{4-biphenyl}(\square)$. Note the change of scale in the insert.

shows the dependence of $1/k'_a$ on the viscosity of ethanol-ethylene glycol mixtures at 0°C as derived from I_M/I_D and measurements of k_{Mf} for compounds of the type $\text{ArCH}_2\text{OCH}_2\text{Ar}$ where $\text{Ar} = \text{phenyl, 1-naphthyl or 4-biphenyl}$ (20). It may be seen that the sensitivity of the conformational mobility to the viscosity of the medium increases dramatically with an increasing volume of the chromophore which has to move through the solution during the conformational transition. In fact, with the bulky diphenyl group even the viscosity of ethanol at 0°C is sufficient to reduce the rate of hindered rotation to one half of the rate estimated for this process in a medium of negligible viscosity.

OTHER LUMINESCENCE STUDIES REFLECTING CONFORMATIONAL MOBILITY

I should like to mention briefly some studies in other laboratories which employed luminescence to study processes depending on the conformational mobility of polymer chains. These are of three types:

(a) If the ends of a flexible chain carry reactive groups whose interaction is diffusion controlled, then the intramolecular reaction, requiring the two chain ends to encounter each other, will be governed by the conformational mobility of the chain. The theory of such processes was first discussed by Wilemski and Fixman (31) and later by Sunagawa and Doi (32). The problem has also been treated by computer simulation (33,34), although this approach was necessarily limited to relatively short chains. An experimental study of such intramolecular diffusion-controlled reactions was first undertaken by Cuniberti and Perico (35) who studied excimer formation in solutions of poly(ethylene oxide) carrying pyrene residues at the two excited ends. This technique lent itself to a study of polymers with up to 1400 atoms in the chain backbone and led to rate constants for chain end encounters which were approximately inversely proportional to the chainlength. Recently, studies

of this type were extended to much longer chains by Usiki *et al* (36). They used monodisperse polystyrene in the DP range 280-3000 with anthryl end groups and followed the delayed fluorescence due to triplet-triplet interaction after a short laser pulse. They concluded that the rate constant for intramolecular chain end encounters is proportional in this range to $(DP)^{-n}$ where $n=0.20\pm 0.05$. This is a much smaller chain length dependence than the $n=2.0\pm 0.5$ predicted by the theoretical treatments.

(b) Diffusion-controlled intermolecular reactions involving end groups of flexible polymer chains are known to be limited by the rate at which the chain ends diffuse to the surface of the molecular coil (37). Thus, such processes also reflect the conformational mobility of polymers. Horie and Mita (38) attached anthracene and benzil moieties to the ends of different monodisperse polystyrene chains and followed the decay of the anthracene phosphorescence after a laser pulse due to quenching by the benzil groups. They found that in the DP range 23-740 the rate constant was proportional to $(DP)^{-0.32}$. In a more recent paper (39) the same technique was used to study the dependence of this rate constant on the solvent power of the medium. It was found that the reaction was slowed down by an increasing polymer solvation for reactions of relatively short chains, while increasing solvation led to increasing rates for reactions of very long chains. This interesting result was interpreted as indicating that for short chains the thermodynamic resistance to the mutual interpenetration of the molecular coils is the dominant effect, while the reaction rate of very long polymers is mostly impeded by topological restraints due to chain entanglement which become more pronounced in poor solvent media.

(c) Fluorimetry has also yielded important data concerning the conformational mobility of a polymer in bulk. Polyvinyl naphthalene (PVN) exhibits excimer fluorescence which may be shown to be due to interactions between neighboring naphthalene residues on the polymer chain. If a film is cast from a solution of polystyrene containing a small concentration of PVN, the PVN is molecularly dispersed in the glassy polystyrene matrix and its I_D/I_M ratio reflects the conformational equilibrium at the temperature at which the film was prepared (40). If the film is now heated to a temperature approaching the glass transition of the polystyrene matrix, the conformational distribution can relax to its equilibrium value and the rate of this change may be monitored by a change in excimer fluorescence intensity. Frank (41) has shown that this process may be followed as much as 25°C below T_g , where the relaxation time is about 160 seconds.

INTERPENETRATION OF FLEXIBLE POLYMER CHAINS IN SOLUTION

If the emission spectrum of a donor chromophore overlaps the absorption spectrum of an acceptor chromophore, nonradiative energy transfer can take place over distances as large as 5 nm. The theory of this process is due to Förster (42) who obtained for the efficiency, E_{eff} , of the energy transfer on the donor-acceptor separation, r , $E_{\text{eff}} = [1 + (r/R_0)^6]^{-1}$ where the sixth power of the characteristic distance R_0 is proportional to the overlap integral. When a solution containing two similar methyl methacrylate copolymers labeled with a donor (naphthyl) and acceptor (anthryl) chromophore, respectively, is irradiated in the donor absorption band, the relative emission intensity from the two chromophores reflects the efficiency of energy transfer, i.e., the probability that the donor and acceptor moieties will approach each other within the critical distance during the excited lifetime of the donor. (Using Förster's theory, spectroscopic properties of this donor-acceptor pair yield $R_0 = 2,3$ nm). The efficiency of energy transfer was found to be considerably lower for the chromophore-labeled polymers than for similar concentrations of donors and acceptors unattached to polymeric carriers. Two effects would be expected to contribute to this difference: First, the much faster diffusion of the small molecules increases the probability of donor-acceptor approach. Secondly, intermolecular energy transfer between polymer chains is impeded by the thermodynamic resistance to mutual interpenetration of the molecular coils in good solvent media.

To distinguish between these two factors, fluorescence spectra were recorded with the donor and acceptor either dissolved in dioxane or incorporated in a poly(methyl methacrylate) film in which diffusion is eliminated (43). The results are shown in Fig. 4. As expected, energy transfer between the small molecules is reduced in the rigid PMMA medium. However, energy transfer between the chromophore-labeled polymers is larger in PMMA than in dioxane, showing that elimination of the excluded volume effect more than compensates for the arrested diffusion.

The same technique may be used to study the interaction of cationic and anionic polymers (44). Here the Coulombic interaction between the two polymeric species would be expected to favor their association and

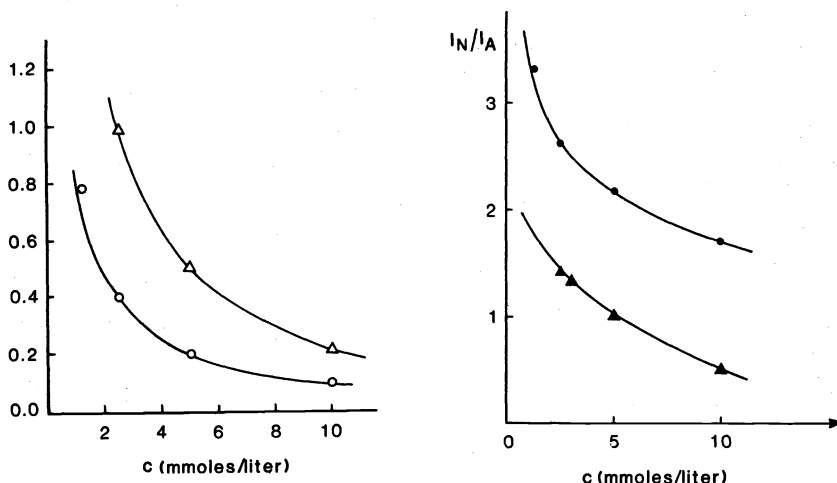


Fig. 4. Ratio of naphthyl and anthryl fluorescence intensities, I_N/I_A , as a function of the chromophore concentration. Circles denote dioxane solutions, triangles solutions in a PMMA matrix. Open symbols refer to systems with free chromophores, filled symbols refer to results with methyl methacrylate copolymers in which 9 mole % of the monomer residues carry chromophore side chains.

energy transfer; this effect should, however, decrease with an increasing ionic strength of the medium. As shown in Fig. 5, experimental results conform to this pattern. In mixtures of cationic and anionic copolymers in which 1.3-1.7 mole % of the monomer residues carry an anthryl or naphthyl chromophore, respectively, I_A/I_N decreases with increasing ionic strength, while energy transfer is independent of electrolyte concentration in solutions of analogous uncharged polymers.

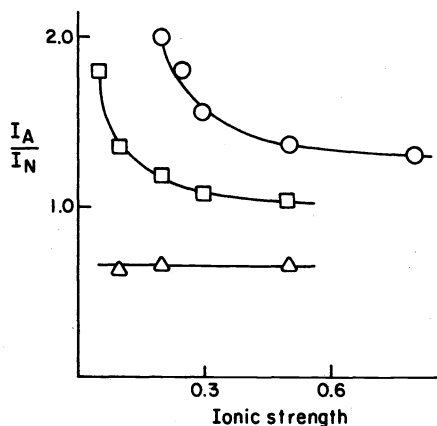


Fig. 5. Dependence of I_A/I_N on ionic strength in solutions containing naphthyl-labeled dimethylacrylamide (DMA) copolymers and anthryl-labeled vinylpyrrolidone (VP) copolymers. (O) DMA copolymers with 14.8 mole % acrylic acid + VP copolymer with 22 mole % dimethylaminoethyl methacrylate. (□) DMA copolymer with 8 mole % acrylic acid + VP copolymer with 13 mole % dimethylaminoethyl methacrylate. (△) Polydimethylacrylamide + polyvinylpyrrolidone.

COMPATIBILITY OF POLYMERS IN BULK

The same principle employed in the preceding section for the study of the mutual interpenetration of chain molecules in solution can be employed to study the compatibility of two polymers in bulk (43,45). It is well known that relatively few polymer pairs form compatible blends (46). Many years ago, Gee pointed out (47) that the mixing of long flexible chain molecules produces a negligible entropy change so that miscibility is only possible if the process is either athermal or exothermic. It would then be expected that if a homopolymer is mixed with a series of copolymers containing a non-polar and a polar component, a one-phase system should be formed when the cohesive energy density of the copolymer matches that of the homopolymer. One of the systems in which such an effect was observed, consisted of mixtures of poly(methyl methacrylate) with styrene-acrylonitrile copolymers (48), where compatible mixtures were observed within a limited range of the copolymer composition. We reinvestigated this system using styrene-acrylonitrile copolymers labeled with donor carbazolyl groups while the poly(methyl methacrylate) was labeled with acceptor anthryl groups. Fig. 6 shows the ratio of the emission intensities of donor and acceptor, I_C/I_A , as a function of copolymer composition (49). The compatibility range is clearly indicated by the pronounced decrease in the I_C/I_A ratio.

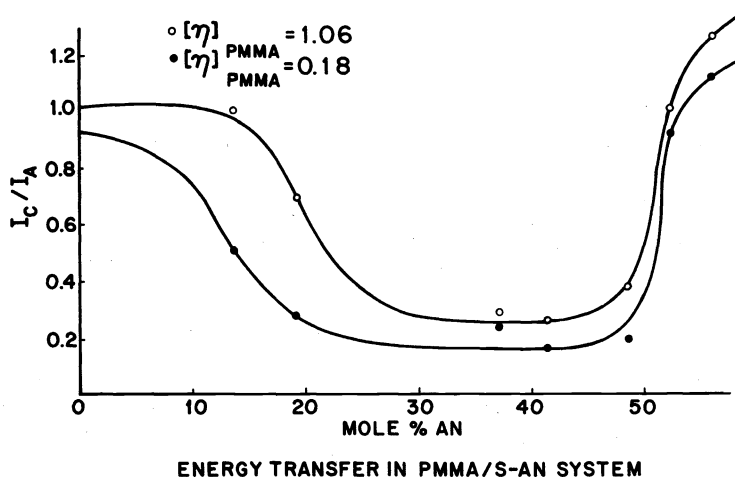


Fig. 6. Ratio of the emission intensity of donor and acceptor in blends of chromophore labeled poly(methyl methacrylate) and styrene-acrylonitrile copolymers as a function of the copolymer composition.

It may also be seen that the intermingling of the chains is favored by a decrease in the molecular weight of the PMMA. Another system which we have investigated (43,45) consisted of blends of poly(methyl methacrylate) and methyl methacrylate copolymers with ethyl or butyl methacrylate. Here the gradual decrease of compatibility with increasing comonomer content was shown to be revealed by the fluorescence technique with much higher sensitivity than by the use of differential scanning calorimetry. An alternative technique employs excimer fluorescence for studying compatibility of polymer blends (50). A polymer such as polyvinyl naphthalene exhibits much lower ratios of monomer and excimer fluorescence in bulk than in dilute solution where the chain is highly expanded. When films were cast from solutions of a series of poly(alkyl methacrylate)s containing a very small concentration of polyvinyl naphthalene (PVN), a plot of I_D/I_M against the difference of the Hildebrand solubility parameters, $\Delta\delta$, of the two polymeric species exhibited a sharp minimum at $\Delta\delta=0$. Thus, when δ of PVN is matched by that of the host matrix, the PVN chain is in true solution.

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