Dissociative Electron Capture of Sulfones and Sulfonates: Matrix Isolation ESR Study

Paul H. Kasai

*Contribution from IBM Almaden Research Center, San Jose, California 95120.
Received December 3, 1990*

Abstract: It is postulated that high sensitivity of poly(olefin sulfones) to electron beam irradiation results from a dissociative electron capture process: $RSO_2R + e^- \rightarrow RSO_2^- + R^{\bullet}$. Dialkyl sulfones RSO_2R and alkyl alkanesulfonates $ROSO_2R$ were trapped in argon matrices together with Na atoms, and electron transfer between them was induced by mild radiation ($\lambda >$ 580 nm). ESR examination of the matrices revealed that all of the sulfones and sulfonates examined (dimethyl sulfone, diethyl sulfone, tetramethylene sulfone, ethyl methanesulfonate, and ethyl trifluoromethanesulfonate) readily captured an electron and dissociated to yield anions RSO_2^- or RSO_3^- and alkyl radicals $R^{\bullet}$. Cyclic tetramethylene sulfone, on capture of an electron, underwent a ring-opening process, yielding $SO_2^-CH_2CH_2CH_2CH_2^{\bullet}$. Ethyl and methyl radicals were generated with equal probability from ethyl methanesulfonate, but only the ethyl radical was generated in the case of ethyl trifluoromethanesulfonate.

Introduction

It has been known that poly(olefin sulfones) **1** undergo rapid chain scission on exposure to high-energy radiation (γ rays and electron beams).[1-3] The polymer **1** is synthesized by a free radical induced chain reaction in a liquid state according to an equilibrium process indicated above. It has been surmised that the primary event in the radiolysis of the polymer is cleavage of weak C–S bonds. The C–S bond scission may then be followed by a de-


1
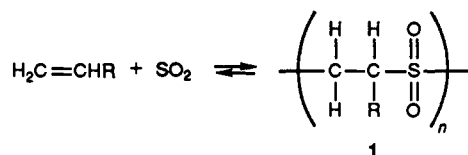
propagation process, resulting in a net G(scission) value of 10 or greater. The extremely high degradation sensitivity of this polymer has been exploited for design of electron beam resists for microlithography as evidence in resist PBS (poly(butene-1 sulfone))[4] or in resist NPR (a mixture of novolac resin and poly(2-methylpentene-1 sulfone)).[5]

The degradation process of poly(olefin sulfones) under high-energy radiation has been extensively examined by O'Donnell and his co-workers.[6-10] Both poly(olefin sulfones) and model monomeric systems such as dialkyl sulfones have been examined. The following reaction sequences have been suggested as the initial step of the observed scission process.
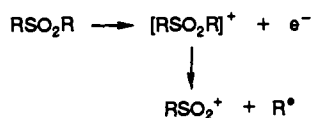
and for poly(olefin sulfones)
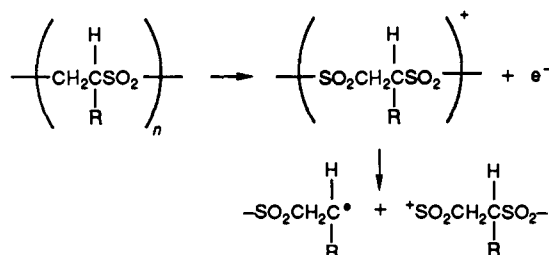
The reaction sequence emanating from the cationic regime was invoked (1) because the ESR spectra of samples irradiated and observed at low temperature (77 K) showed mainly those of alkyl radicals and little due to sulfonyl radicals $RSO_2^{\bullet}$ and (2) in order to account for isomerization observed on olefin monomers released in radiolysis of the polymer. The ESR spectrum of sulfonyl radicals appeared on warming of the samples irradiated at low temperature.

When a material is exposed to a beam of high-energy electrons or γ ray radiation, a swarm of thermal energy electrons are generated through cascading ionization processes. When the energy of the primary beam is $\sim$10 KeV, for example, the total number of secondary electrons generated is estimated to be 10^3 times that of the primary electrons.[11] The electron affinity of the SO_2 molecule has been determined as 2.2 eV.[12] Taking cognizance of the high electron affinity of the SO_2 sector, we postulated that the primary step in radiolysis of alkyl sulfones and poly(olefin sulfones) might be a dissociative electron-capture process ($RSO_2R + e^- \rightarrow RSO_2^- + R^{\bullet}$).

The technique of cocondensing metal atoms of low ionization potential (e.g., Na atoms) and molecules of interest in an argon matrix and effecting electron transfer between them by mild radiation, thus generating *chemically isolated* metal cations and either the molecular anions or the products of the dissociative electron-capture process, has been demonstrated and described many years ago.[13] The mild radiation required for the electron transfer ($\lambda >$ 580 nm in the case of Na atoms) ensures that the electrons being captured are of thermal energy, and neither the starting acceptors nor the products of the capture process are subjected to photoradiation of consequence. This report presents the results of an ESR study of such matrix-isolated, electron-transfer reactions conducted in order to demonstrate the propensity of dialkyl sulfones and, by inference, that of poly(olefin sulfones) to undergo a dissociative electron-capture process. The electron affinity of the SO_3 molecule is also high (1.7 eV).[14] The elec-

(1) Ayscough, P. B.; Ivin, K. J.; O'Donnell, J. H. *Trans. Faraday Soc.* **1965**, *61*, 1110.
(2) Bowden, M. J.; Thompson, L. F. *J. Appl. Polym. Sci.* **1973**, *17*, 3211.
(3) Bowden, M. J. *J. Polym. Sci. Polym. Chem. Ed.* **1974**, *12*, 499.
(4) Thompson, L. F.; Bowden, M. J. *J. Electrochem. Soc.* **1973**, *120*, 1722.
(5) Bowden, M. J.; Thomspon, L. F.; Farenholtz, S. R.; Doerries, E. M. *J. Electrochem. Soc.* **1981**, *128*, 1304.
(6) Brown, J. R.; O'Donnell, J. H. *Macromolecules* **1970**, *3*, 265.
(7) Brown, J. R.; O'Donnell, J. H. *Macromolecules* **1972**, *5*, 109.
(8) Bowmer, T. N.; O'Donnell, J. H.; Wells, P. R. *Makromol. Chem. Rapid Commun.* **1980**, *1*, 1.
(9) Bowmer, T. N.; O'Donnell, J. H. *Radiat. Phys. Chem.* **1981**, *17*, 177.
(10) Bowmer, T. N.; O'Donnell, J. H. *Polymer* **1981**, *22*, 71.

(11) Parikh, M. *J. Chem. Phys.* **1980**, *73*, 93.
(12) Fukuda, E. K.; McIver, Jr., R. T. *J. Chem. Phys.* **1982**, *77*, 4942.
(13) Kasai, P. H. *Acc. Chem. Res.* **1971**, *4*, 329.

tron-capture reaction of alkyl alkanesulfonates $ROSO_2R$ was hence also investigated.

The study revealed that all the sulfones and sulfonates examined (dimethyl sulfone, diethyl sulfone, tetramethylene sulfone, ethyl methanesulfonate, and ethyl trifluoromethanesulfonate) readily captured an electron and dissociated to yield anions RSO_2^- or RSO_3^- and alkyl radicals $R^\bullet$. Cyclic tetramethylene sulfone, on capture of an electron, underwent a ring-opening process, yielding $SO_2^-CH_2CH_2CH_2CH_2^\bullet$. Ethyl and methyl radicals were generated with equal probability from ethyl methanesulfonate, but only the ethyl radical was generated in the case of ethyl trifluoromethanesulfonate.

Experimental Section

The liquid helium cryostat-ESR spectrometer system, which would enable trapping of vaporized metal atoms in inert gas matrices and examination of the resulting matrices by ESR, has been described earlier.[13] The technique of trapping Na atoms as electron donors and molecules of interest as acceptors and inducing electron transfer between them by mild radiation ($\lambda > 580$ nm), thus generating cations and products of the electron-capture process effectively isolated in inert gas matrices, has also been described.[13]

In the present series of experiments, the Na atoms were generated from a resistively heated stainless steel tube (250 °C). Dimethyl sulfone, diethyl sulfone, and tetramethylene sulfone were introduced from a glass sample reservoir heated to ~100 °C. Ethyl methanesulfonate was introduced from a glass sample reservoir at room temperature. Ethyl trifluoromethanesulfonate was premixed (2%) with argon gas. All the sulfone and sulfonate chemicals examined were obtained from Aldrich Chemical Co.

The ESR spectrometer used was an IBM Instruments Model 200D, X-band spectrometer, and the microwave frequency locked to the sample cavity was typically 9.420 GHz. For photoirradiation of the matrix, a high-pressure Xe–Hg lamp (Oriel, 1 kW unit) was used. The light beam was passed through a water filter and a broad-band interference filter (600 ± 50 nm) and focused on the cold finger ~40 cm away.

Spectra and Assignments

Dimethyl Sulfone. An argon matrix in which the Na atoms and dimethyl sulfone molecules had been cocondensed showed, without any photoirradiation, strong ESR signals due to methyl radicals (Figure 1a, a quartet with $A = 23.3$ G) and weak signals due to Na atoms. The ESR spectrum of Na atoms $(3s^1)$ is a sharp, widely spaced quartet due to a large, isotropic hyperfine interaction $(A = 330$ G) with the ^{23}Na nucleus. When the matrix was irradiated with yellow light (600 ± 50 nm), the Na signals disappeared completely with corresponding increase in the intensity of the methyl radical quartet. The result is attributed to electron transfer from Na to dimethyl sulfone and spontaneous dissociation of the resulting anions.

$$Na + CH_3SO_2CH_3 \longrightarrow Na^+ + [CH_3SO_2CH_3]^-$$

$$\downarrow$$

$$CH_3SO_2^- + CH_3^\bullet$$
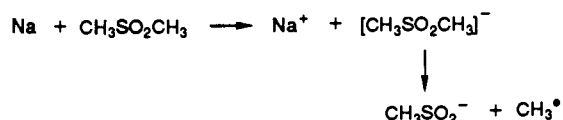
The transfer is apparently very facile for this pair. The majority of the reaction occurred through collision in space (as the two beams merged above the cold finger) and in the fluid surface layer of the matrix during deposition.

The ESR spectrum of the SO_2^- anion is known. It had been detected in γ-irradiated sodium dithionite[15] and in γ-irradiated potassium metabisulfite.[16] Its g tensor has been reported as $g_1 = 2.0103$, $g_2 = 2.0055$, and $g_3 = 2.0018$. In order to ascertain the electron affinity of SO_2 itself and to observe the ESR spectrum of SO_2^- anions generated in an argon matrix, a separate experiment was performed in which Na atoms were trapped in an argon matrix
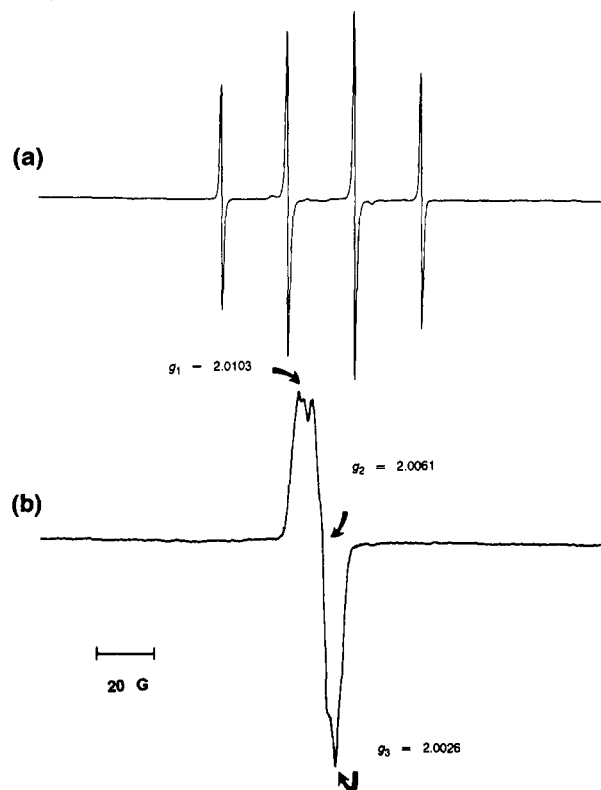
Figure 1. (a) ESR spectrum observed from an argon matrix in which Na and dimethyl sulfone had been cocondensed. The sharp quartet is due to methyl radicals, and the signals due to Na atoms are outside the range shown. (b) ESR spectrum observed from an argon matrix in which Na and SO_2 had been cocondensed. The broad singlet signal is due the SO_2^- anion; its g tensor is assessed as indicated.

containing SO_2 (1%). The resulting matrix showed, without photoirradiation, weak signals due to Na atoms and an extremely strong singlet signal near the position corresponding to $g = 2.00$ (Figure 1b). The latter signal has a pattern due to radicals having an orthorhombic g tensor as indicated and is hence assigned to the SO_2^- anion radical. The complete absence of the signal due to SO_2^- in Figure 1a is significant since it shows that the $CH_3SO_2^-$ anion does not dissociate further in the dissociative electron-capture process of dimethyl sulfone.

Diethyl Sulfone. When Na atoms and diethyl sulfone were cocondensed in an argon matrix, extremely strong ESR signals were observed near and centered about the $g = 2.00$ position (Figure 2a). As in the case of dimethyl sulfone, these signals were observed without photoirradiation, and the signals due to Na atoms were weak. Upon irradiation with yellow light (600 ± 50 nm), the Na atom signals disappeared completely and the intensity of the signals in Figure 2a increased further. The spectral pattern seen in Figure 2a is that of ethyl radicals. The ethyl radical had been generated in argon matrices (by photolysis of HI in the presence of ethylene), and its ESR spectrum had been analyzed by McDowell et al.[17] Figure 2b is a computer-simulated powder pattern of ethyl radicals based on the following parameters: $g_\parallel = g_\perp = 2.0023$, $A_\parallel = 30.0$, and $A_\perp = 19.0$ G for the two α protons, and $A_\parallel = 29.6$ and $A_\perp = 26.0$ G for the three β protons. These parameters represent the situation where the methyl group is rotating and the two α protons are exchanging their positions rapidly. In stimulating the spectrum, it was further assumed that the three protons in the methyl group assumed the nuclear spin state of $I_{total} = {}^3/_2$ only reflecting the temperature of the matrix (~4 K). It has been shown that the lowest rotational level of the methyl internal rotor can assume only the nuclear spin state of $I_{total} = {}^3/_2$.[18] The hfc (hyperfine coupling) constants determined
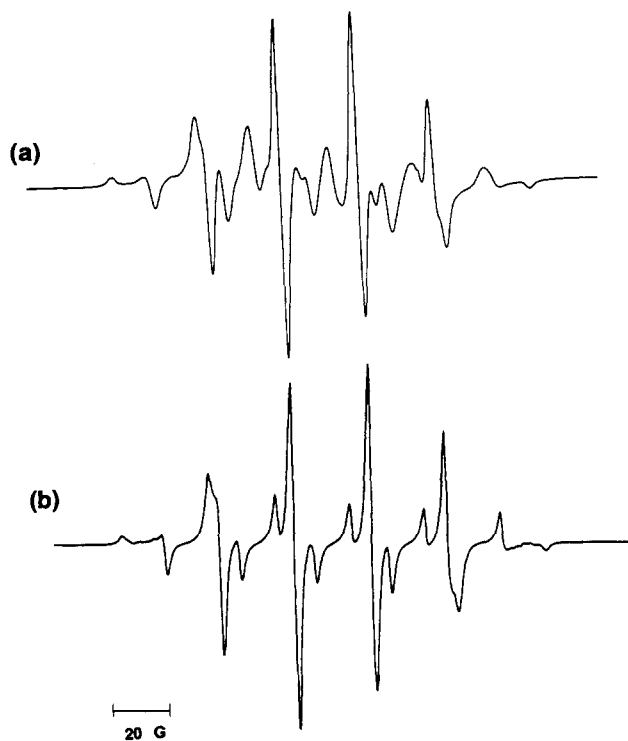
(14) Rothe, E. W.; Tang, W. Y.; Reck, G. P. *J. Chem. Phys.* **1975,** *62,* 3829.

(15) Clark, H. C.; Horsfield, A.; Symons, M. C. R. *J. Chem. Soc.* **1961,** 7.

(16) Atkins, P. W.; Horsfield, A.; Symons, M. C. R. *J. Chem. Soc.* **1964,** 5220.

(17) McDowell, C. A.; Raghunathan, P.; Shimokoshi, K. *J. Chem. Phys.* **1973,** *58,* 114.

(18) Heller, C. *J. Chem. Phys.* **1962,** *36,* 175.

Figure 2. (a) ESR spectrum observed from an argon matrix in which Na and diethyl sulfone had been cocondensed. (b) Computer-simulated spectrum of ethyl radicals based on the parameter given in the text.
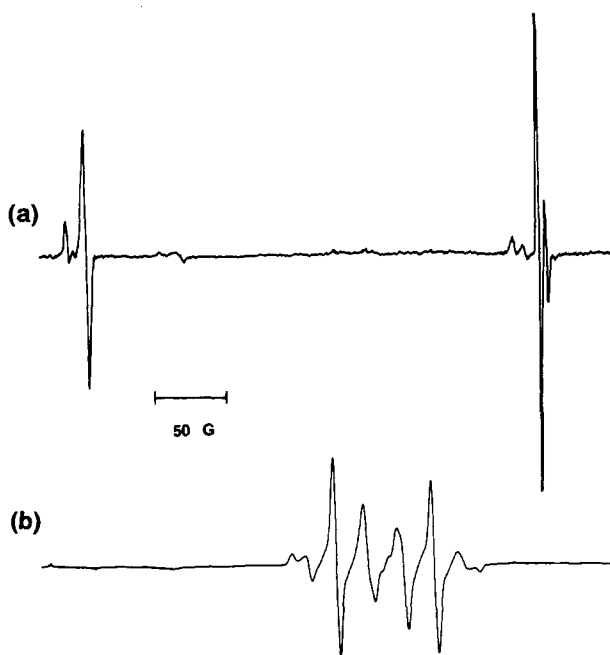
Figure 3. ESR spectra of an argon matrix containing Na atoms and tetramethylene sulfone. (a) Observed before photoirradiation; the signals seen at both ends are inner components of the quartet due to Na atoms. (b) Observed after photoirradiation with yellow light (600 ± 50 nm).

here are very similar to those reported earlier.[17] Generation of ethyl radicals in the present matrix must be ascribed to dissociative electron capture of diethyl sulfone.

Tetramethylene Sulfone (Sulfolane). An argon matrix in which Na atoms and tetramethylene sulfone had been cocondensed showed, prior to photoirradiation, only the signals due to Na atoms. Figure 3a and b shows the ESR spectra (the section encompassing the inner pair components of the Na atom quartet) of the matrix observed before and after irradiation with yellow light (600 ± 50 nm) for 8 min.
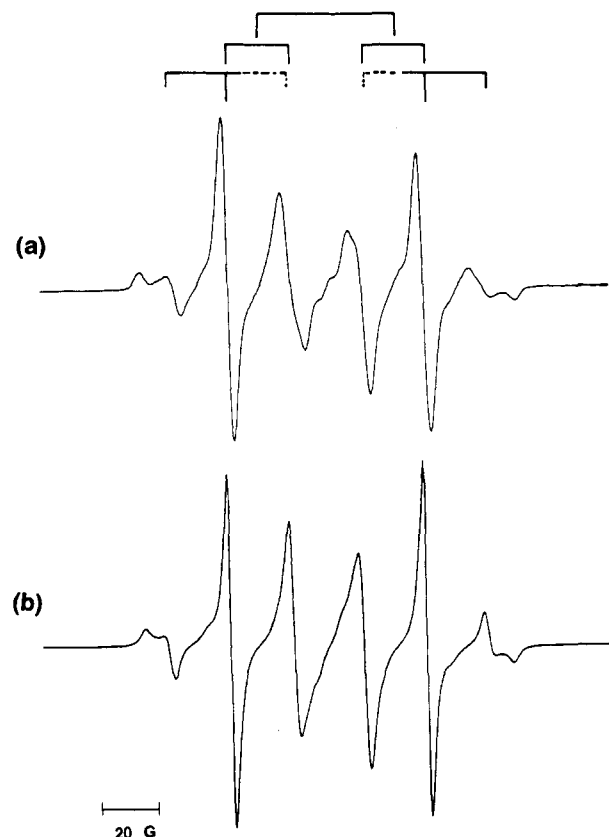

Figure 4. (a) ESR signals photoinduced in the Na/tetramethylene sulfone/Ar system shown in an expanded scale. A doublet of doublet of triplet pattern was recognized as indicated. (b) Computer-simulated spectrum of $SO_2^-CH_2CH_2CH_2CH_2^\bullet$ anions based on the parameters given in the text.

Disappearance of the Na signals upon photoirradiation and simultaneous appearance of a multiplet pattern centered about the $g = 2.00$ position are attributed to the Na to sulfone electron transfer and spontaneous ring rupture of the resulting anions as shown below.
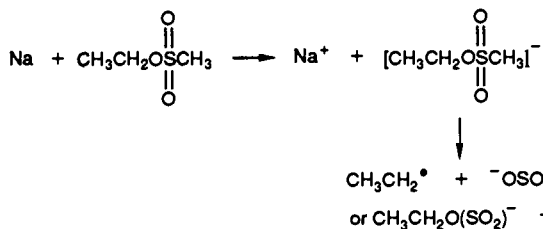

$$SO_2^- CH_2CH_2CH_2CH_2^\bullet$$
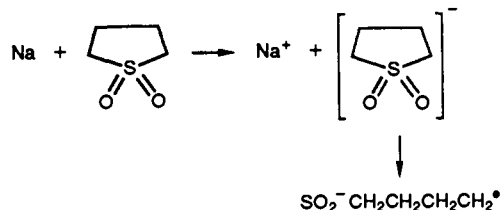
The anion of the ring-rupture form is a primary alkyl radical, the ESR spectral pattern of which is determined essentially by the hyperfine interactions with the two α protons and the two β protons. The photoinduced spectrum is shown expanded in Figure 4a. It is recognized to possess a doublet of doublet of triplet pattern as indicated. The spacings of the triplet ($\sim$20 G) and the anisotropic line shape of its outer components are characteristics of the hyperfine interaction with two rapidly exchanging α protons. The essentially isotropic doublet of doublet splittings of 46 and 22 G are hence assigned to the two β protons. Again the absence of the ESR signal due to SO_2^- signifies that the sulfolane anion of the ring-rupture form does not dissociate further in the postulated electron-transfer process. Further analysis of the spectrum of the anion is presented in the Discussion.

Ethyl Methanesulfonate. An argon matrix in which Na atoms and ethyl methanesulfonate had been cocondensed showed, without photoirradiation, strong ESR signals due to methyl and ethyl radicals. The signals due to Na atoms were weak and disappeared completely on irradiation with yellow light (600 ± 50 nm). Figure 5a shows the observed spectrum of the alkyl radical section. It is compared with the simulated spectrum (Figure 5b) based on

(a)

(b)

|——————|
20 G
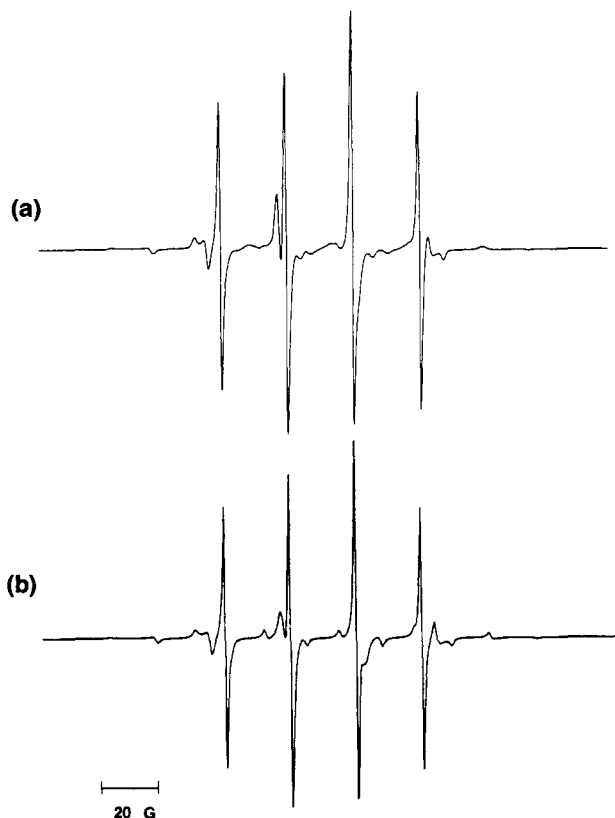
Figure 5. (a) ESR spectrum observed from an argon matrix in which Na atoms and ethyl methanesulfonate had been cocondensed. (b) Computer-simulated spectral pattern based on the presence of methyl and ethyl radicals in equal abundance.

the presence of methyl and ethyl radicals in equal abundance. The ESR signal of the SO_3^- anion is also known. It is a singlet signal at $g = 2.0036$.[19] There was no evidence of SO_3^- in the observed spectrum (Figure 5a). We thus conclude that, in the dissociative electron capture of ethyl methanesulfonate, either the ethyl radical or the methyl radical could dissociate with equal probability, and in either case, the resulting anion does not dissociate further.

$$Na + CH_3CH_2OSCH_3 \longrightarrow Na^+ + [CH_3CH_2OSCH_3]^-$$

$$\downarrow$$

$$CH_3CH_2^\bullet + {}^-OSO_2CH_3$$
$$or \ CH_3CH_2O(SO_2)^- + CH_3^\bullet$$

Ethyl Trifluoromethanesulfonate. An argon matrix in which Na atoms and ethyl trifluoromethanesulfonate had been cocondensed showed, without photoirradiation, strong ESR signals due to ethyl radicals and weak signals due to Na atoms. Upon irradiation with yellow light (600 ± 50 nm), the Na atom signals disappeared completely and the signals due to ethyl radicals increased. No other signal was observed. The result is ascribed to the Na to sulfonate electron transfer and spontaneous dissociation of the resulting anions as given below.

$$Na + CH_3CH_2OSCF_3 \longrightarrow Na^+ + [CH_3CH_2OSCF_3]^-$$

$$\downarrow$$

$$CH_3CH_2^\bullet + {}^-OSO_2CF_3$$
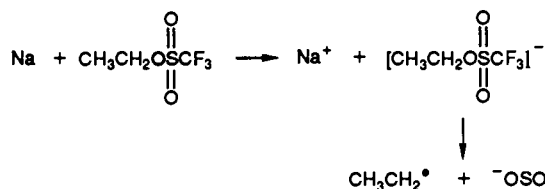
(19) Chantry, G. W.; Horsfield, A.; Morton, J. R.; Whiffen, D. H. *Mol. Phys.* **1962**, *5*, 233.
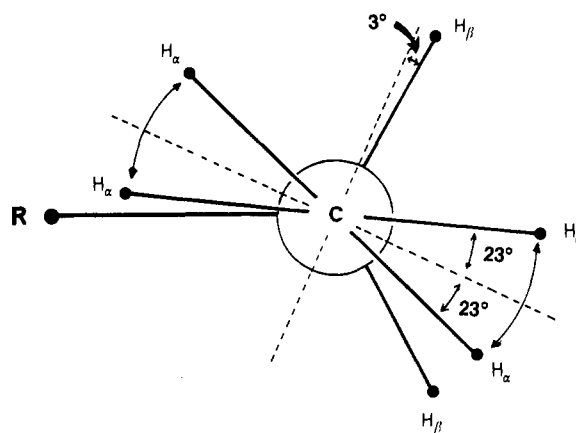
Figure 6. Conformation of $SO_2^-CH_2CH_2CH_2CH_2^\bullet$ anion radicals. The H_α-C_α-H_α plane oscillates as shown.

Discussion

Conformation of $SO_2^-CH_2CH_2CH_2CH_2$. The hfc tensors of α and β protons of an alkyl radical are well-understood. The hfc tensor of an α proton is rather anisotropic and may be given as follows.[20,21]

$$A_\parallel = 12 \ G \qquad A_{\perp\parallel} = 22 \ G \qquad A_{\perp\perp} = 35 \ G$$

Here, $A_\parallel$ is the diagonal element of the tensor in the direction parallel to the C–H bond, and $A_{\perp\parallel}$, for example, is the diagonal element perpendicular to the C–H bond but parallel to the p_π orbital of the α carbon. Thus, the orientations of the hfc tensors of two α protons of an alkyl radical differ by $\sim120°$ about the p_π orbital, and the triplet pattern with a prominent central component would not be observed unless the two protons exchange their positions rapidly (>40 MHz). The hfc tensor of a β proton is essentially isotropic and is determined by the dihedral angle θ of the C_β–H_β bond relative to the p_π orbital at the α carbon.[22]

$$A(H_\beta) = B_0 + B \cos^2 \theta$$

It has been empirically determined that $B_0 = 4$ G and $B = 50$ G. Thus, as the dihedral angles of the two β protons differ by $\sim120°$, their hfc constants would generally be different.

As stated earlier, the triplet pattern recognized in Figure 4a is characteristic of the hfc interaction with two rapidly exchanging α protons. It can be readily shown that the β proton splittings of 46 and 22 G are also incompatible with a static model. A situation was hence assumed whereby the H_α-C_α-H_α plane oscillates $\pm\Delta$ degrees centered about a certain orientation. The hfc constant of one of the β protons would be the average of the two values given by the two dihedral angles $\eta \pm \Delta$, while the hfc constant of the second β proton would be the average of those corresponding to the dihedral angles $\eta + 2\pi/3 \pm \Delta$. Here, η would be the average dihedral angle of the first C_β–H_β bond. Analysis of the observed coupling constants of 46 and 22 G according to this scheme yields $\eta = 3°$ and $\Delta = 23°$. The Newman diagram of the resulting conformation is shown in Figure 6.

Figure 4b is a computer-simulated spectrum based on the model α and β proton hfc tensors, exchange of the α protons at 300 MHz, and oscillation of the H_α-$C\alpha$-H_α plane as defined in Figure 6 at the rate of 340 MHz.[23] The revealed conformation and dynamics of the radical terminus are, not unexpectedly, very similar to those concluded earlier for the n-propyl radical by Adrian et al[24] and more recently for the trimethylene oxide anion of the ring-ruptured form.[25]

(20) McConnell, H. M.; Strathdee, J. *Mol. Phys.* **1959**, *2*, 129.
(21) Kasai, P. H. *J. Am. Chem. Soc.* **1972**, *94*, 5950.
(22) Morton, J. R. *Chem. Rev.* **1964**, *64*, 453.
(23) A usual ESR powder pattern program (ref 21) was modified to include the effect of exchanging protons by the method of modified Bloch equation.
(24) Adrian, F. J.; Cochran, E. L.; Bowers, V. A. *J. Chem. Phys.* **1973**, *59*, 3946.

Dissociative Electron Capture. The experimental results obtained in the present study clearly establish the propensity of sulfone and sulfonate molecules to undergo a dissociative electron-capture process. Poly(olefin sulfones), having a higher proportion of the sulfone group, are expected to be more vulnerable to such a process. The present result does not rule out the mechanism suggested earlier for the radiolysis of these materials; it does give credence to the new postulate that the primary step in the radiolysis of these materials might be dissociative electron capture. Absence of the ESR signals due to sulfonyl radicals $RSO_2^{\bullet}$ in samples irradiated at low temperature may be ascribed to the process where the sulfonyl end remains as an anion (RSO_2^-). Emergence of the ESR signals due to sulfonyl radicals on warming may be attributed to thermal release of trapped electrons and/or holes and resulting electron hole recombination. Isomerization observed in the olefinic monomers released in the radiolysis of poly(olefin sulfones) may have occurred during depropagation of polymer chains having ionized sectors.

The Na to sulfone (or sulfonate) electron transfer actually effected was so facile that the majority of the reaction occurred

during deposition through collision in space or in a fluid surface layer of the developing matrix. Tetramethylene sulfone was an exception in that no electron transfer was observed prior to photoirradiation. The exception, we believe, is due to a lower sample pressure possible from tetramethylene sulfone and to a steric factor unfavorable for the collision process.

In an effort to gain further insight to the process involved, molecular orbital calculations (EHT) were performed. The calculations showed that the LUMO (lowest unoccupied orbital) in the cases of dimethyl sulfone and tetramethylene sulfone would be an antibonding σ orbital confined to the CSC sector. For methyl methanesulfonate and methyl trifluoromethanesulfonate, the predicted LUMO was an antibonding σ orbital confined to the CO sector. In methyl methanesulfonate, the second lowest unoccupied orbital was shown to be an antibonding σ orbital confined to the CS sector. The corresponding orbital in the methyl trifluoromethanesulfonate was much higher in energy as expected. The predicted energy levels of these orbitals are such that the stable anions (of the original structure) would not be formed. The dissociative electron capture of sulfones and sulfonates must then occur through repulsive negative ion resonance states, and the point of scission is determined by the electronegativity of the SO_2 or SO_3 moiety and weakness of the C–S and C–O bonds.

(25) Kasai, P. H. *J. Am. Chem. Soc.* **1990**, *112*, 4313.

Photoregulation of Papain Activity through Anchoring Photochromic Azo Groups to the Enzyme Backbone

Itamar Willner,* Shai Rubin, and Azalia Riklin

Contribution from the Department of Organic Chemistry, The Hebrew Univeristy of Jerusalem, Jerusalem 91904, Israel. Received January 22, 1990. Revised Manuscript Received August 21, 1990

Abstract: The enzyme papain has been chemically modified by 4-carboxyazobenzene (**1**), 3-carboxyazobenzene, (**2**), and 2-carboxyazobenzene, (**3**). The activities of the modified enzymes relative to native papain and the extent of loading by the azobenzene groups have been determined. *trans*-1–papain exhibits reversible photochromic properties, and upon illumination ($\lambda = 320$ nm), interconversion to *cis*-1–papain occurs. Further illumination of *cis*-1–papain ($\lambda > 400$ nm) regenerates *trans*-1–papain. The biocatalyst *trans*-1–papain is 2.75-fold more active than *cis*-1–papain toward hydrolysis of N^{α}-benzoyl-DL-arginine-4-nitroanilide (BAPNA; **4**). The difference in activities of the trans/cis photochromic enzyme allows photoregulation of the hydrolytic process. Kinetic analyses reveal that the difference in activities of the two photochromic forms of 1–papain originates from poorer binding properties of *cis*-1–papain toward the substrate, as compared to *trans*-1–papain. The biocatalyst *trans*-1–papain is immobilized into Alginate beads, and cyclic photoregulated hydrolysis of BAPNA is effected.

Photoregulation of enzyme activities provides a general approach to utilize biocatalysts as light-controlled "on–off" switching systems. Potential applications of photoregulated enzymes include their use as light signal amplifiers, information storage devices, and sensing assemblies.[1,2] Photoregulation of enzymes has been previously described by using photochromic materials that act as inhibitors for the enzyme active site specifically in one of their photochromic configurations.[3,4] Also, chemical derivatization of the enzyme active site by photochromic substituents is capable of blocking the biocatalyst activity in one of the photochromic structures.[5] Other approaches to photoregulate enzymes involve chemical modification of the biocatalyst active site with photoactive

groups deactivating the enzyme, but upon illumination, the blocking group is released and consequently the biocatalyst activity is restored.[6] In a single report,[7] chemical modification of the protein backbone of enzymes by the spiropyran photochromic material led to photoregulation of a few enzymes. The photoregulative activities of these enzymes have been attributed to alterations in hydrophilicity–hydrophobicity of the protein microenvironments resulting from the neutral–zwitterionic photochromic forms of spiropyran. Nevertheless, comparison of the chemically modified enzyme activities to the native catalysts is scarce. Also, in all of the reported photoregulated biocatalytic assemblies no cyclic "on–off" switching performance is documented. Here, we wish to report on photoregulation of papain by anchoring azobenzene photochromic groups to the protein backbone. Azobenzene undergoes photoinduced trans to cis

(1) Hug, D. *Photochem. Photobiol. Rev.* **1978**, *3*, 1–33.
(2) Martinek, K.; Berezin, I. V. *Photochem. Photobiol.* **1979**, *29*, 637–649.
(3) Bieth, J.; Vratsanos, S. M.; Wassermann, N. H.; Erlanger, B. F. *Proc. Natl. Acad. Sci. U.S.A.* **1969**, *64*, 1103.
(4) Wainberg, M. A.; Erlanger, B. F. *Biochemistry* **1971**, *10*, 3816.
(5) (a) Erlanger, B. F.; Wassermann, N. H.; Copper, A. G.; Monk, R. J. *Eur. J. Biochem.* **1976**, *61*, 287. (b) Montagnoli, G.; Balestreri, E.; Nannicini, L.; Bellucci, A.; Bracaloni, M. *Int. J. Pept. Protein Res.* **1978**, *11*, 28.

(6) Turner, A. D.; Izzo, S. V.; Rozakis, G. W.; Porter, N. A. *J. Am. Chem. Soc.* **1987**, *109*, 1274–1275.
(7) Aizawa, M.; Namba, K.; Suzuki, S. *Arch. Biochem. Biophys.* **1977**, *182*, 305–310.