1308 © *1989 The Chemical Society of Japan* *Bull. Chem. Soc. Jpn.*, ***62***, *1308—1312 (1989)* [Vol. 62, No. 4

A Time-Resolved CIDEP Study of the Laser Photoreduction of Pyruvic Acid by Lactic Acid

Marjory T. Craw, M. Catherine Depew, Frank J. Adrian,† and Jeffrey K. S. Wan*
Department of Chemistry, Queen's University, Kingston, Ontario, K7L 3N6 Canada

(Received July 29, 1988)

In addition to the well-known initial phototriplet polarization, unusual Chemically Induced Dynamic Electron Polarization (CIDEP) behavior in solutions of neat pyruvic acid/lactic acid has been attributed to ST_- Radical Pair Mechanism (RPM) polarization in combination with absorptive initial Triplet Mechamism (TM) polarization and ST_0 RPM polarization. Significant ST_- RPM contributions to such carbon centered radicals with small hyperfine splittings have rarely been observer in solution at room temperature.

Pyruvic acid has been utilized quite extensively in this laboratory in studies of free radical reactions with compounds of biological interest including vitamin C, vitamin E, and several quinones.[1–3] The first demonstration of enhanced absorptive triplet mechanism polarization was evidenced in the photoreduction of pyruvic acid;[1] subsequently photolysis of pyruvic acid in glassy ethanol matrices was utilized to demonstrate the concept of spin conservation in energy transfer processes in randomly oriented matrices.[4]

While the pyruvic acid system was an earlier model in our studies to establish the simultaneous contributions of both the initial Triplet Mechanism and the additional ST_0 Radical Pair mechamism in both CIDEP and CIDNP[1] phenomena, the possible operation of the ST− radical pair polarization had eluded our attention until our recent understanding of this mechanism in a more general, but separate chemical system.[5] (The designations ST− and ST_0 refer to polarizations that arise from the interaction or mixing of the singlet and T− or T_0 triplet energy sublevels of the radical pair during the period of separation of the members of the radical pair in which the energies of the two sublevels are nearly degenerate.) In this report, we show the unusual polarization patterns in a series of photoreduction studies of neat pyruvic acid by lactic acid. The complex CIDEP behavior of these systems is due to the substantial and simultaneous contributions of ST− RPM polarization in addition to the expected ST_0 contributions. The significant ST− contribution in the present pyruvic/lactic acid system is mainly due to the very strong molecular interaction between the pair leading to a very high molecular viscosity.

Experimental

Pyruvic acid, lactic acid, and hexafluoro-2-propanol were obtained from Aldrich. Solution viscosities were measured at 25 °C using a falling ball viscosimeter. The CIDEP experiments were conducted using a modified Varian E3 spectrometer without field modulation, as described previously.[6] The solutions were irradiated in situ with a Lambda Physik EMG101-MSC XeCl excimer laser at 308 nm. The path length of the cell used was 0.8 mm in order to reduce any inner filter effects for light absorption.

Results and Discussion

Photolysis of pyruvic acid/lactic acid solutions results in the initial formation of a pair of identical ketyl radicals as shown below.

$$CH_3COCOOH + CH_3CHOHCOOH \longrightarrow 2\,CH_3\dot{C}OHCOOH$$

The time-resolved CIDEP spectrum of a 1 : 1 v/v mixture of pyruvic acid (PA), and lactic acid (LA) in 50% ethanol is shown in Fig. 1a. There is very good agreement between the experimental spectrum and a spectrum simulated for 40% TM and 60% ST_0 polariza-

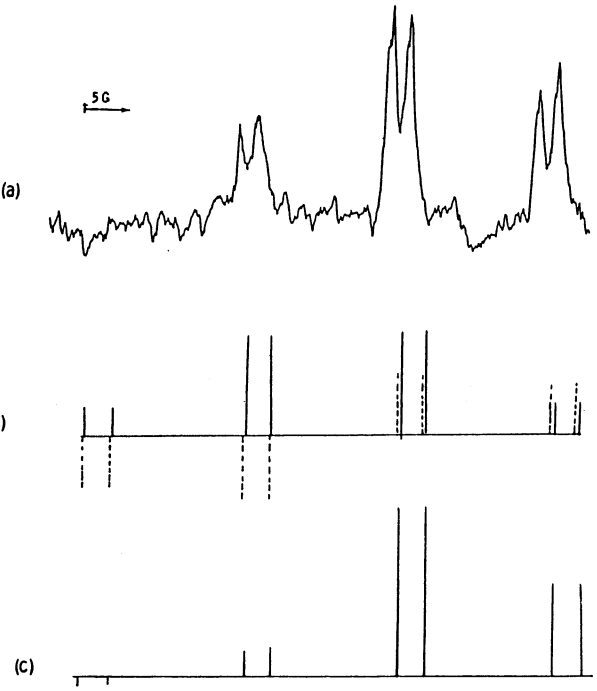


Fig. 1. (a) Time-resolved magnetic resonance spectrum of pyruvate radicals formed following photolysis of a 1 : 1 v/v PA/LA mixture in 50% ethanol. (b) Expected intensity distribution and phase of TM (——) polarization and ST_0 (----) (right shifted for observation). (c) Simulated CIDEP spectrum for 40% TM and 60% ST_0 radical pair mechanism polarization.

---

† Permanent address: Milton S. Eisenhower Research Center, Applied Physics Laboratory, the Johns Hopkins University, Laurel, Maryland 20707, U. S. A.

tion (Fig. 1b and 1c).

However, when neat solutions of PA and LA are mixed the resulting solution appears to have a substantially increased viscosity and the relative intensity distribution and phase of the hyperfine lines in the CIDEP spectrum of the ketyl radical changes rather dramatically. Figure 2a—c represents typical spectra observed for neat PA/LA mixtures as a function of relative concentrations of each component, measured at 3 μs after the laser flash. These spectra show that as the pyruvic acid concentration increases the LF/HF ratio also becomes larger and more negative, thus suggesting some additional contribution from ST− mixing.

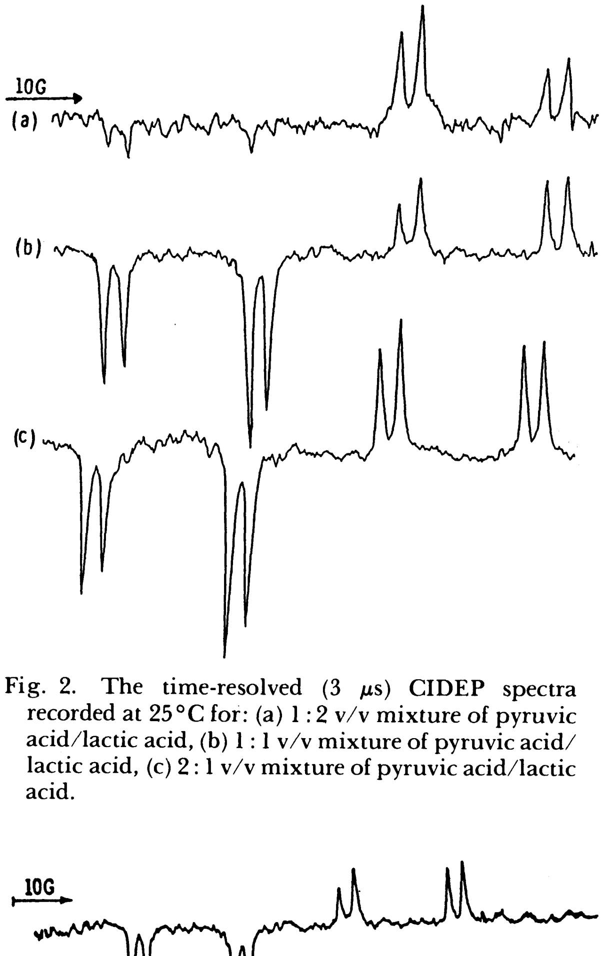


Fig. 2. The time-resolved (3 μs) CIDEP spectra recorded at 25 °C for: (a) 1 : 2 v/v mixture of pyruvic acid/lactic acid, (b) 1 : 1 v/v mixture of pyruvic acid/lactic acid, (c) 2 : 1 v/v mixture of pyruvic acid/lactic acid.

While several examples of a rather large hyperfine induced ST− contributions to polarization spectra have been published,[5,8] the development of "viscosity" induced ST− polarization has not been well-characterized. The CIDEP spectra of 1 : 1 v/v PA/LA at varying temperatures as shown in Fig. 3a—c clearly exhibit a dramatic temperature effect. At 25 °C the relative ratios of the LF/HF lines indicate the presence of an ST− contribution to the observed polarization spectrum. On lowering the temperature the relative intensity of the high field lines diminishes until at −15 °C the phase of these hyperfine lines changes entirely to emission. These observations are consistent with an increase in the ST− contribution to the CIDEP spectrum as the molecular viscosity of the system increases.

Additional insight into the effects of viscosity on this system was obtained by examining the effects of a variety of solvents with a wide range of viscosities from glycerol/water mixtures to nonviscous solvents such as ethanol, hexafluoro-2-propanol, benzene, and formic acid. In neat 1 : 1 v/v mixtures of PA/LA and in solutions containing 50% ethanol or glycerol/water mixtures the viscosity of each solution was measured at 30 °C, ranging from 13 cP (1P=0.1 Pa s) for a solution of 50% ethanol/PA/LA to 73 cP for neat PA/LA system, to 580 cP for a 1 : 1 mixture of PA/LA and glycerol. The time-resolved CIDEP spectra observed

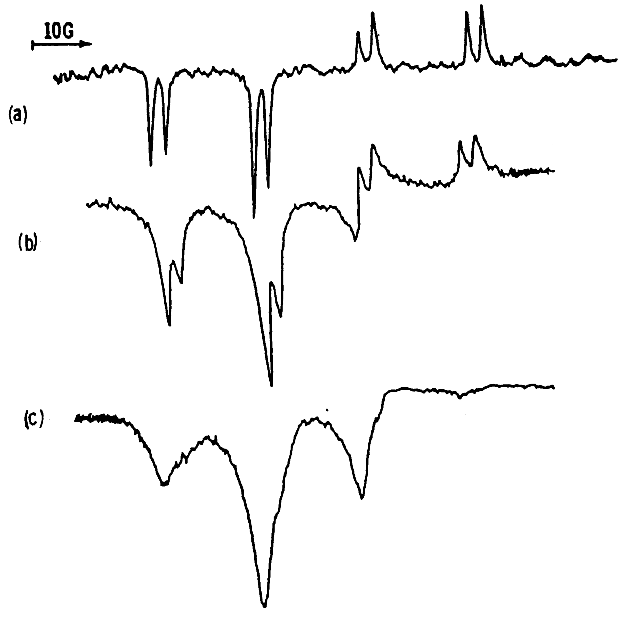


Fig. 3. The CIDEP spectra for a 1 : 1 v/v mixture of pyruvic acid/lactic acid at (a) 25 °C, (b) −2 °C, (c) −15 °C.

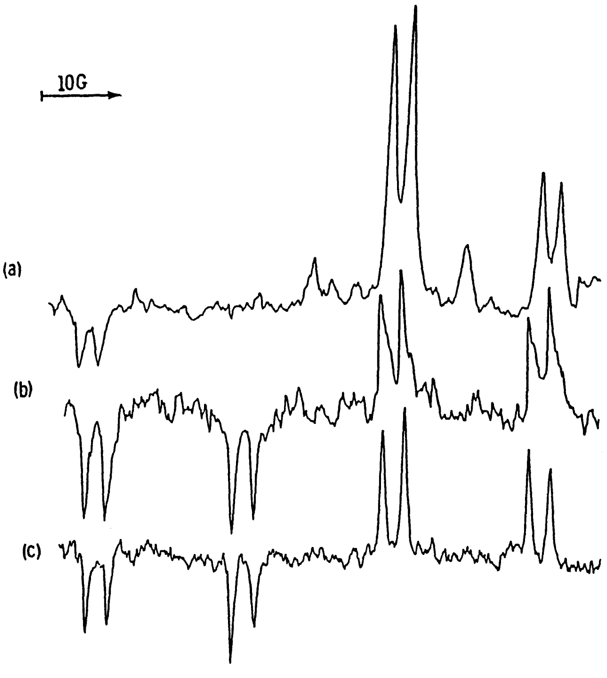


Fig. 4. The time-resolved (3 μs) CIDEP spectra for 1 : 1 PA/LA in 50%. (a) Glycerol, (b) hexafluoroisopropanol, (c) benzene.

for pyruvate radicals under various solvent conditions are shown in Fig. 2a—c. There is no apparent direct correlation between solvent viscosity and the development of ST− polarization for pyruvate radicals, since the most viscous solvent-glycerol-resulted in less ST− contribution than benzene or hexafluoro-2-propanol, or neat PA/LA itself (Fig. 3a).

Pyruvic acid molecules under certain circumstances may dimerize or trimerize, presumably through their carboxyl moieties.[9] This unique property of pyruvic acid may lead to a strong interaction between it and its reduced form, lactic acid. Such a strong molecular interaction may occur both in the ground state and in the radical pair state. Thus ST− mixing may be enhanced when the residence time of an initially formed radical pair in the level crossing region is increased according to[11]

$$\rho_{\mathrm{ST-}} = (\pi A^2/4g\,\mu_{\mathrm{B}}{}^{\mathrm{H}})\,(r_{\mathrm{c}}/2\lambda D) \tag{1}$$

where A is the difference in hyperfine frequencies of the pair and D is the diffusion coefficient. We have used the Smoluchowski equation and the previously published rate constants for the disproportionation of pyruvate radicals in various solvents[7] to calculate D as a function of viscosity,

$$K = 4\pi\sigma\rho DN/1000 \tag{2}$$

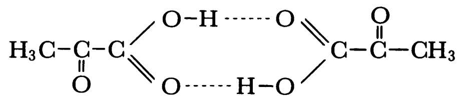
where $K=1/2$ the termination rate constant, σ the spin statistical factor for free radical reaction, N is Avogadro's number and ρ the distance of closest approach estimated to be in the region of 6 Å.

The data are presented in Table 1; the assumptions made here suggest that D for pyruvate radicals in 2—4×10^{-6} M pyruvic acid (1 M=1 mol dm^{-3}) in ethanol (viscosity 1.2 cP) is aproximately 10E−5 cm^2 s^{-1}, not unreasonable for organic radicals in solution.[12] As the viscosity is increased the diffusion of the radicals in a pair would be expected to be very restricted leading to a significant contribution of ST− polarization. While a qualitative analysis suggests that the ST− mechanism becomes more important as the viscosity is increased in a particular solvent, several of the experimentally observed spectra indicate that viscosity arguments alone cannot account for the magnitude of the ST− polarization contributions in defferent solvents. Clearly in solvents such as glycerol and glycerol/water systems the CIDEP spectrum shows contributions from ST$_0$, TM, and ST− polarizations. However, while the development of ST− polarization for C centered radicals has been observed previously at low temperature[13] and in systems where the solvent viscosity was high,[14] in both cases the ratio of ST−/ST$_0$ polarization was relatively small.

Table 1. Estimated Diffusion Coefficients for Pyruvate Radicals as a Function of Solvent Viscosity[7]

Solvent	D	Viscosity
	cm^2 s^{-1}	cP
Ethanol	1.5×10^{-5}	1.2
2-Propanol	7.05×10^{-6}	2.3
1-Butanol	4.23×10^{-6}	3.9
2-Methyl-1-propanol	4.75×10^{-6}	3.9
Benzyl alcohol	4.4×10^{-6}	4.1

As mentioned above pyruvic acid is known to form a polymeric structure, dimer or trimer, in nonpolar solvents.[15] This dimer presumably arises via a hydrogen bonding type of interaction similar to that previously reported for acetic acid.[9] Therefore, it is not surprising that solvent interactions that affect this hydrogen bonding should result in changes in the observed time-resolved spectra. Pyruvic acid has a pK_a of 2.39, while that for lactic acid is 3.83,[16] therefore when ethanol, pK_a=10, is added to a PA/LA acid, it is possible that the concentrations of the protonated forms of the acids are reduced which may limit the extent of such a hydrogen bonding interaction. Comparing the spectra for PA/LA in 50% ethanol with those observed for PA/LA in 50% formic acid and for PA/LA/50% $(CF_3)_2COH$ in which the viscosities of the solutions are similar, a much larger contribution from ST− polarization is found in both of the latter cases. Both $(CF_3)_2CHOH$ have pK_a values in the region of 4—6,[16] therefore in these systems the hydrogen bonding interaction may still be significant, as a result of the acidity of the solvent. This may be further substantiated by the PA/ethyl lactate experiments in which the apparent ST− contribution decreases markedly when the acid functionality of LA is esterified.



When glycerol is used as solvent, the viscosity of the system increases but pK_a for glycerol is ≃14.[16] Therefore the contribution of ST− polarization predicted on the basis of viscosity comparisons may be somewhat lower than expected since in this solvent system the extent of hydrogen bonding and thus the degree of "internal" restricted diffusion may itself be reduced as a result of deprotonation of the acid functionalities. Furthermore, the observed substantial ST− polarization in benzene solutions are consistent with the reduced deffusion expected as a result of the known preference of PA to exist in a polymeric structure in this nonpolar solvent.[15] Also the observed increase in the relative contribution of ST− mixing with increasing concentrations of pyruvic acid is consistent with an increase in the extent of association of pyruvic acid molecules as the concentration relative to LA is increased.

Obviously the complexity of both the physicochemical interactions between the reagants and the solvents in this system and the presence of three contributing polarization mechanisms does not render

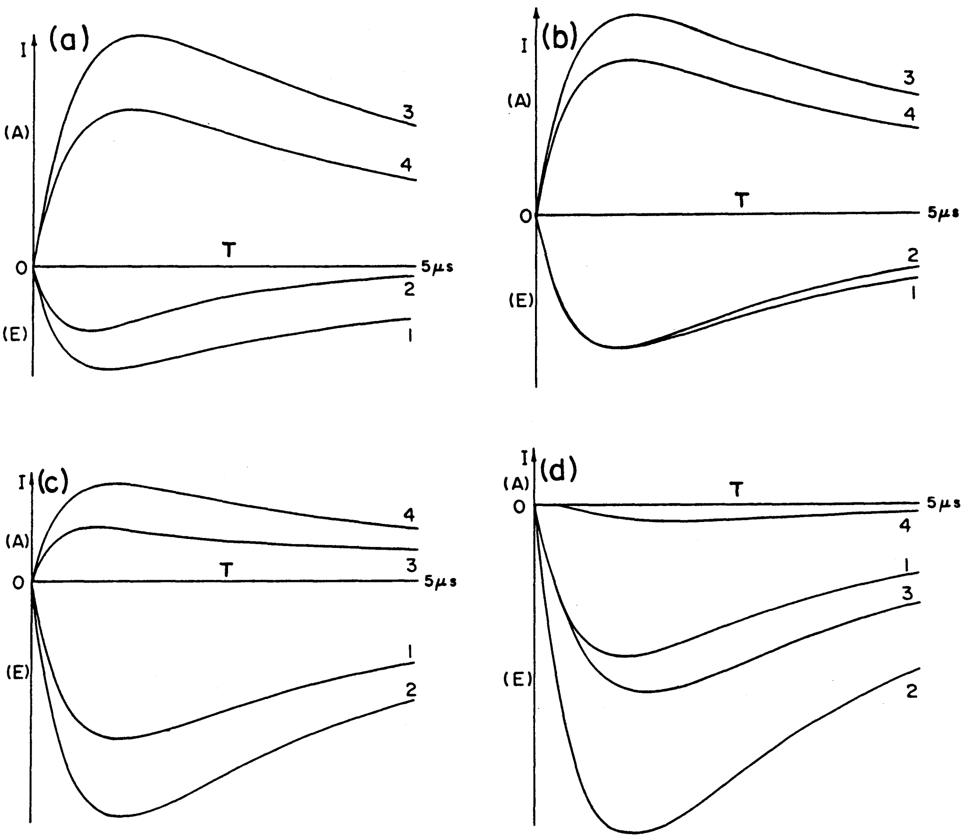


Fig. 5. The calculated line intensities and phases for RPM polarization of pyruvate ketyl radical at various values of D.
(a) $D=10^{-5}$ cm^{2}s^{-1}, $\rho_{ST_0}=0.033$, $\rho_{ST_-}=10^{-4}$, (b) $D=10^{-6}$ cm^{2}s^{-1}, $\rho_{ST_0}=0.063$, $\rho_{ST_-}=10^{-3}$, (c) $D=10^{-7}$ cm^{2}s^{-1}, $\rho_{ST_0}=0.103$, $\rho_{ST_-}=0.01$, (d) $D=10^{-8}$ cm^{2}s^{-1}, $\rho_{ST_0}=0.151$, $\rho_{ST_-}=0.1$.

quantitative examination possible. However an analysis of the variation of the relative hyperfine line intensities and phases with decreasing deffusion between the members of the radical pair which contributes differing relative ST_0 and $ST-$ components to the expected polarization shows some interesting results. Computer simulation of the effects of decreasing D from 10^{-5} to 10^{-8} cm^2 s^{-1} is shown in Fig. 5. The relative probabilities of ST_0 to $ST-$ mixing (ρ_{SY_0}/ρ_{ST-}) vary from 330 to 1.5 on going from Fig. 5a to 5d. It is interesting to note the close correlation of line intensity changes with decreasing D value expected in several of the experimental situations; for example, in 50% ethanol the time-resolved spectrum (Fig. 1a) is consistent with the pattern predicted in 5a (with an additional absorptive contribution due to the TM), while in 1 : 2 PA/LA (Fig. 2a) the pattern is quite similar to that shown in 5b. The line intensity changes exhibited in Fig. 3a—c as a result of decreased diffusion due to lowering of the temperature are in good agreement with the changes predicted in Fig. 5c and d.

Finally it is interesting to note the biological role of pyruvic acid and the environment that these molecules exist in in vivo, which can also be studied by CIDEP. We hope to extend this study to other systems in which the possibility of molecular interactions influencing the development of CIDEP may be of importance.

References

1) K. Y. Choo and J. K. S. Wan, *J. Am. Chem. Soc.*, **97**, 7127 (1975).

2) M. C. Depew, B. B. Adeleke, and J. K. S. Wan, *Can. J. Chem.*, **59**, 2708 (1981).

3) J. K. S. Wan, *J. Photochem.*, **17**, 517 (1981).

4) D. Weir and J. K. S. Wan, *J. Am. Chem. Soc.*, **106**, 427 (1984); K. Obi and T. Imamura, *Rev. Chem. Intermed.*, **7**, 225 (1986).

5) T. J. Burkey, J. Lusztyk, K. U. Ingold, J. K. S. Wan, and F. J. Adrian, *J. Phys. Chem.*, **89**, 4268 (1985).

6) J. W. DeBoer, T. Y. C. Chan Chung, and J. K. S. Wan, *Can. J. Chem.*, **59**, 2708 (1981); E. Kam, M. T. Craw, M. C. Depew, and J. K. S. Wan, *J. Mag. Reson.*, **67**, 556 (1986).

7) P. B. Ayscough and R. C. Sealy, *J. Chem. Soc., Perkin Trans. 2*, **1973**, 543.

8) K. Akiyama, private communication.

9) R. W. Taft and M. J. Kamlet, *J. Chem. Soc., Perkin Trans. 2*, **1979**, 1723.

10) S. Yamauchi, K. Tominaga, and N. Hirota, *J. Phys. Chem.*, **90**, 236 (1986).

11) F. J. Adrian and L. Monchick, *J. Chem. Phys.*, **71**, 2600 (1979).

12) T. J. Burkey, D. Griller, D. A. Lindsey, and J. C. Scaiano, *J. Am. Chem. Soc.*, **106**, 1983 (1984).

13) C. D. Buckley and K. A. McLauchlan, *Chem. Phys. Lett.*, **137**, 86 (1987).

14) A. D. Trifunac, D. J. Nelson, and C. Mottley, *J. Magn. Res.*, **30**, 263 (1978).

15) P. A. Leermakers and G. F. Vesley, *J. Am. Chem. Soc.*, **85**, 3776 (1963).

16) R. Stewart, "The Proton: Applications to Organic Chemistry," Academic Press, Inc., New York (1984), Vol. 46, Chap. 2; P. Ballinger and F. A. Long, *J. Am. Chem. Soc.*, **82**, 795 (1960); "Ionization Constants of Organic Acids in Aqueous Solutions-IUPAC-," Pergammon Press, IUPAC (1979), Vol. 23, pp. 41, 45.

17) C. D. Buckley, D. A. Hunter, P. J. Hore, and K. A. MacLauchlan, *Chem. Phys. Lett.*, **135**, 307 (1987).