

sets and including polarization functions, previously considered to be infallible in the prediction of the qualitative aspects of molecular structure, fail to produce any deviation from linearity in the heavy-atom chain.^{8,9} Following the publication of the experimental evidence that propadienone has a C_s rather than C_{2v} structure, quantum chemists have shown that when electron correlation is included either by perturbation¹⁰ techniques or in multi-configurational-SCF calculations^{11,12} then the predicted energy minimum for propadienone occurs for an unsymmetrical structure similar to the experimentally determined one reported here. Brown and Dittman¹¹ showed that the kinked geometry is primarily attributable to a contribution to the ground state from the lowest energy doubly excited electronic configuration. Farnell and Radom¹⁰ have pointed out that the flatness of the ν_{12} potential function at the Hartree-Fock level makes it feasible for any conflicting influence arising from electron correlation to manifest itself in a resultant double-minimum potential function.

Taylor¹² employed the recently developed CASSCF method with an augmented double- ζ plus polarization basis set to obtain a fully optimized equilibrium structure. The heavy-atom angles were predicted to be $\angle C_1C_2C_3 = 139.5^\circ$ and $\angle OC_1C_2 = 165.5^\circ$.

A number of ketene homologues and related molecules have been studied by spectroscopic methods, and it appears that unusually flat potentials for bending vibrations analogous to the ν_{12} mode of propadienone are the norm rather than exceptional within this group; the methylene wag force constants for ketene ($C-H_2:C:O$)¹³ and diazomethane ($CH_2:N:N$)^{14,15} are respectively one-third and one-fifth of that for ethylene; the ν_{15} (out of

heavy-atom plane) mode of cyclopropylidene methanone ($\overline{CH_2 \bullet CH_2 \bullet C:C:O}$)¹⁶ and the ν_7 mode of carbon suboxide ($O:C:C:O$)¹⁷ each have very shallow potentials with small central barriers rising to the vicinity of the vibrational ground-state energy; the ν_{15} mode of butatrienone ($CH_2:C:C:O$)¹⁸ has a shallow single minimum potential.

In each of these molecules, the ease of deformation may be attributable qualitatively to the influence of a canonical form containing $=C_2-C_1 \equiv O^+$ or in the case of diazomethane the isoelectronic $=C-N^+ \equiv N$ analogous to II for propadienone. The influence appears usually merely to flatten the bending potential without distorting the equilibrium structure, but in a few cases, stabilization of the bent form may be sufficient to lower the symmetry of the equilibrium structure. To date, propadienone is the only experimentally observed example for which substantial potential minima (in comparison with zero-point vibrational energy) develop for the deformed structure. From the viewpoint of ab initio molecular orbital calculations, propadienone is a unique case in which the inclusion of electron correlation is essential for the correct prediction of the symmetry of the equilibrium structure.

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Registry No. $H_2CCC^{18}O$, 79048-71-8; $H_2CC^{13}CO$, 96445-82-8; $H_2^{13}CCCCO$, 79048-72-9; $HDCCCO$, 64918-70-3; H_2CCCO , 61244-93-7.

Supplementary Material Available: Tables of ground-state rotational transitions for isotopic variants of propadienone and correlation matrices from the least-squares fit (5 pages). Ordering information is given on any current masthead page.

(8) Radom, L. *Aust. J. Chem.* **1978**, *31*, 1-9.

(9) Kormonicki, A.; Dykstra, C. E.; Vincent, M. A.; Radom, L. *J. Am. Chem. Soc.* **1981**, *103*, 1652-1656.

(10) Farnell, L.; Radom, L. *Chem. Phys. Lett.* **1982**, *91*, 373-377.

(11) Brown, R. D.; Dittman, R. G. *Chem. Phys.* **1984**, *83*, 77-82.

(12) Taylor, P. R. *J. Compt. Chem.*, in press.

(13) Moore, C. B.; Pimentel, G. C. *J. Chem. Phys.* **1963**, *38*, 2816-2829.

(14) Moore, C. B.; Pimentel, G. C. *J. Chem. Phys.* **1964**, *40*, 329-341.

(15) Moore, C. B.; Pimentel, G. C. *J. Chem. Phys.* **1964**, *40*, 342-355.

(16) Brown, R. D.; Godfrey, P. D.; Kleibömer, B.; Champion, R.; Elmes, P. S. *J. Am. Chem. Soc.* **1984**, *106*, 7715-7718.

(17) Lozes, R. L.; Sabin, J. R.; Oddershede, J. *J. Mol. Spectrosc.* **1981**, *86*, 357-364.

(18) Brown, R. D.; Ball, M. J.; Champion, R.; Godfrey, P. D.; McNaughton, D.; Rodler, M., to be published.

The Microwave Spectrum and Structure of Tricarbon Monoxide

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Contribution from the Department of Chemistry, Monash University, Clayton, Victoria, Australia 3168. Received January 18, 1985

Abstract: The microwave spectra of tricarbon monoxide isotopically substituted with ^{13}C or ^{18}O at each of its four atoms in turn are reported. From the derived rotational constants the full substitution geometry of this linear molecule is derived, viz., $r_s(C_1O) = 115.0$ pm, $r_s(C_1C_2) = 130.6$ pm, $r_s(C_2C_3) = 125.4$ pm. Discrepancies with ab initio molecular orbital calculations (MP3/6-31G*) are somewhat larger than usual, perhaps because of the effect of a low-frequency bending vibration. From observations of l -doubling transitions we estimate the bending frequency to be about 150 cm^{-1} , reasonably near the theoretical prediction of 200 cm^{-1} . The oxide was generated from a more convenient precursor—fumaroyl dichloride. The preparations of required isotopically enriched versions of this compound are described.

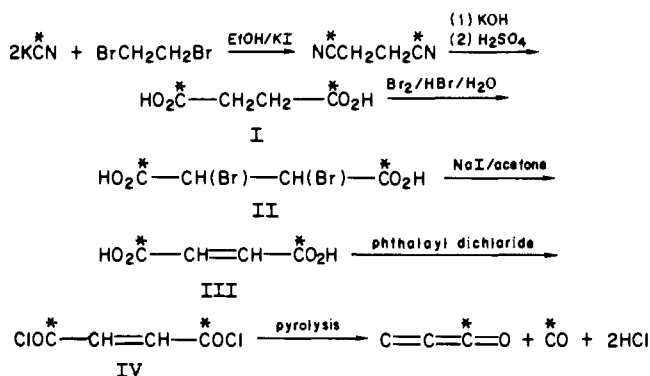
We have recently reported the generation of C_3O by pyrolysis, its identification being based on observations of six lines in the microwave spectrum of the common isotopic species,¹ the line frequencies being in close accord with predictions based on ab initio molecular orbital calculations on the properties of C_3O .²

We now report a more extensive study of the microwave spectrum of C_3O , including all monoisotopically substituted versions of the molecule. We also describe an alternative pyrolysis for generating C_3O from fumaroyl dichloride ((*E*)-2-butenediol dichloride) that has proved more convenient both for isotopic synthesis and for spectroscopic observations.

(1) Brown, R. D.; Eastwood, F. W.; Elmes, P. S.; Godfrey, P. D. *J. Am. Chem. Soc.* **1983**, *105*, 6496-6497.

(2) Brown, R. D.; Rice, E. H. *J. Am. Chem. Soc.* **1984**, *106*, 6475-6478.

Scheme I



Experimental Section

Preparation of Precursors. [$^{18}\text{O}_2$]Fumaroyl Dichloride. Maleic anhydride (1.0 g, 0.01 mol) and [^{18}O]water (26% ^{18}O , 1.4 g, 0.07 mol) were heated in a sealed tube at 100 °C for 24 h. The excess water was recovered (17% ^{18}O by mass spectrometry) and the maleic acid was dried under vacuum (60 °C (0.001 mm)). The intensities of the parent ion mass peaks at m/z 124 ($\text{C}_4\text{H}_4^{18}\text{O}_4$, <1%), 122 ($\text{C}_4\text{H}_4^{18}\text{O}_3^{16}\text{O}_1$, 2%), 120 ($\text{C}_4\text{H}_4^{18}\text{O}_2^{16}\text{O}_2$, 14%), 118 ($\text{C}_4\text{H}_4^{18}\text{O}_1^{16}\text{O}_3$, 41%), and 116 ($\text{C}_4\text{H}_4^{16}\text{O}_4$, 43%) (and the ^{18}O concentration of the recovered water) indicate complete isotopic exchange of all oxygen atoms. [$^{18}\text{O}_2$]Fumaroyl dichloride was prepared from [$^{18}\text{O}_2$]maleic acid (1.16 g, 0.01 mol) and phthaloyl dichloride (4.6 g, 0.02 mol) in the presence of anhydrous zinc chloride at 130 °C.³ At the end of the reaction the [$^{18}\text{O}_2$]fumaroyl dichloride was vacuum distilled (60 °C (0.001 mm)) to a cooled (-196 °C) receiver. No further purification of the sample was necessary. A comparison of the intensities of sets of mass spectral lines at m/z 123 ($\text{C}_4\text{H}_2^{18}\text{O}_2^{37}\text{Cl}$, 0.4%), 121 ($\text{C}_4\text{H}_2^{18}\text{O}^{16}\text{O}^{37}\text{Cl} + \text{C}_4\text{H}_2^{18}\text{O}_2^{35}\text{Cl}$, 6.4%), 119 ($\text{C}_4\text{H}_2^{16}\text{O}_2^{37}\text{Cl} + \text{C}_4\text{H}_2^{18}\text{O}^{16}\text{O}^{35}\text{Cl}$, 32%), and 117 ($\text{C}_4\text{H}_2^{16}\text{O}_2^{35}\text{Cl}$, 50.1%) or at m/z 93 ($\text{C}_3\text{H}_2^{18}\text{O}^{37}\text{Cl}$, 5%), 91 ($\text{C}_3\text{H}_2^{16}\text{O}^{37}\text{Cl} + \text{C}_3\text{H}_2^{18}\text{O}^{35}\text{Cl}$, 47.4%), and 89 ($\text{C}_3\text{H}_2^{16}\text{O}^{35}\text{Cl}$, 100%) indicated an ^{18}O isotopic enrichment for C_3O of about 13%.

[1,4- $^{13}\text{C}_2$]Fumaroyl Dichloride. The above compound was prepared by the sequence of reactions shown in Scheme I with potassium [^{13}C]cyanide (90%, ^{13}C).

[1,4- $^{13}\text{C}_2$]Succinic acid (I) was obtained by alkaline hydrolysis of [1,4- $^{13}\text{C}_2$]succinonitrile prepared by the reaction of potassium [^{13}C]cyanide (1.0 g, 15 mmol) with excess 1,2-dibromoethane (2.62 g, 14 mmol).⁴ The beige-colored product (I) (0.7 g, 77%), mp 178–180 °C (lit. mp 188 °C)⁵ was used without further purification. ^1H NMR (acetone- d_6) δ 2.6 (d, $J = 2$ Hz, 4 H, CH_2), 8.55 (br s, 2 H, OH); MS, m/z 120 (M^+ , <1), 102 ($\text{M}^+ - \text{H}_2\text{O}$, 63), 75 (45), 74 (50), 73 (25), 57 (20), 56 (100), 46 (80).

[1,4- $^{13}\text{C}_2$]-(*R,S*)-2,3-Dibromosuccinic acid (II) was prepared from [1,4- $^{13}\text{C}_2$]succinic acid (0.6 g, 5 mmol) as previously described.⁶ The product (II) (0.91 g, 65%) was isolated as a white powder after sublimation (195 °C (3 \times 10⁻² mm)). ^1H NMR (acetone- d_6) δ 4.65 (d, $J_{\text{C}_1\text{H}_1} = 2$ Hz, 2 H, CH), 8.7 (br s, 2 H, OH).

[1,4- $^{13}\text{C}_2$]Fumaric acid (III) was obtained as a cream-colored solid (0.30 g, 80%), mp 299 °C (lit.⁵ mp 300–302 °C), by debromination of [1,4- $^{13}\text{C}_2$]-(*R,S*)-2,3-dibromosuccinic acid (0.91 g, 3.3 mmol).⁶ ^1H NMR (acetone- d_6) δ 6.77 (t, $J_{\text{C}_1\text{H}_1} \approx J_{\text{C}_2\text{H}_2} = 4.85$ Hz), 6.77 (d, $J = 4.85$ Hz due to 18% [^{13}C]fumaric acid). MS, m/z 118 (M^+ , 20), 117 (3), 100 (100), 89 (25), 83 (20), 72 (20), 54 (40), 46 (75).

[1,4- $^{13}\text{C}_2$]Fumaroyl dichloride (IV) was prepared from [1,4- $^{13}\text{C}_2$]fumaric acid (III) (0.3 g, 2.6 mmol) and phthaloyl dichloride (1.1 g, 5.4 mmol) as outlined above. A pink-colored liquid (0.3 g, 76%) was obtained and pyrolyzed without further purification. MS, m/z 154 (M^+ , absent), 119 ($\text{M}^+ - \text{Cl}$, 65), 90 (100), 64 (25), 55 (38), 42 (24).

[2- ^{13}C]Fumaroyl Dichloride. The procedure used for the synthesis of [2- ^{13}C]fumaroyl dichloride was essentially the same as in Scheme I except that the sequence began with [2- ^{13}C]succinic acid (90% ^{13}C , 0.3 g, 2.5 mmol) supplied as a beige-colored powder, mp 179–81 °C (lit.⁶ mp

188 °C), by Stohler Isotope Chemicals. MS, m/z 119 (M^+ , absent), 101 ($\text{M}^+ - \text{H}_2\text{O}$, 60), 75 (60), 74 (60), 73 (25), 57 (43), 56 (96), 45 (100).

[2- ^{13}C]-(*R,S*)-2,3-Dibromosuccinic acid (0.49 g, 70%): ^1H NMR (acetone- d_6) δ 4.73 (s, 10% unlabeled dibromosuccinic acid), 4.73 (8 lines, 90% [2- ^{13}C]dibromosuccinic acid, $J_{\text{C}_1\text{H}_1} \approx 166$ Hz, $J_{\text{C}_2\text{H}_2} \approx 8$ Hz, $J_{\text{H}_1\text{H}_2} \approx 12$ Hz), 10.9 (br s, OH).

[2- ^{13}C]Fumaric acid mp 285 °C dec; MS, m/z 117 (M^+ , 18), 116 (2), 100 (20), 99 (100), 89 (18), 82 (18), 72 (18), 54 (30), 45 (75).

[2- ^{13}C]Fumaroyl dichloride was obtained as a pink-colored liquid (0.2 g, 84%) from [2- ^{13}C]fumaric acid (0.18 g, 1.6 mmol) and phthaloyl dichloride (0.69 g, 3.4 mmol). MS, m/z 153 (M^+ , absent), 118 ($\text{M}^+ - \text{Cl}$, 70), 90 (100), 63 (30), 55 (35), 42 (25).

5,5'-Bis(2,2-dimethyl-4,6-dioxo-1,3-dioxanylidene). Repeated attempts to prepare this compound according to the literature method⁷ gave erratic results. More reliable results and improved yields were obtained by adding 5-bromo-2,2-dimethyl-1,3-dioxan-4,6-dione (0.74 g, 3.3 mmol) to a solution of 5,5-dibromo-2,2-dimethyl-1,3-dioxan-4,6-dione (1 g, 3.3 mmol) in dry DMF (10 mL) which had been standing for 1 week in a well-illuminated place. Dissolution of the 5-bromo-2,2-dimethyl-1,3-dioxan-4,6-dione was quickly followed by precipitation of the required dimer (0.48 g, 57%): mp 230 °C dec; MS, m/z 284 (M^+ , absent), 182 (6), 154 (18), 101 (12), 80 (C_4O_2 , 30), 58 (35), 52 (C_3O , 5), 43 (100).

Spectrometers and Pyrolyses. For measurements up to 60 GHz a conventional microwave spectrometer with 30-kHz Stark modulation was used. Phase-locked OKI Klystrons served as microwave sources. The 1 m long G-band sized cell, which was kept at room temperature, was immediately followed by a liquid nitrogen cooled trap to allow a high flow rate. Pyrolyses were conducted by flowing the vapor of the appropriate precursor through a silica tube (30 cm length, 2 cm i.d.) in an electric furnace. Temperatures were measured with a thermocouple on the external wall of the tube. The emerging gases were pumped through the Stark cell at pressures of 0.2 to 1.5 Pa.

Some measurements were made at higher frequencies with a tone-burst modulated spectrometer, the sources being harmonic generators driven by phase-locked OKI Klystrons (60V12 or 70V11).

For accurate measurements of line-center frequencies the spectrometers were interfaced to a Varian V75 computer system, lines being repetitively scanned and averaged. The center frequencies were derived by computer fitting of a Lorentzian curve by a least-squares technique.

With use of fumaroyl dichloride as starting material the maximum yield of tricarbon monoxide was obtained with an oven temperature of around 950 °C. In order to get strong lines of C_3O it was essential to condition the surface of the brass Stark cell. This was achieved by pyrolysing 3–4 g fumaroyl dichloride vapor at 1000 °C and 2–3 Pa. This yielded a dark brown coating on the quartz tube and the Stark cell without noticeably affecting the microwave properties of the cell. The use of a new silica tube instead of a conditioned one did not influence the intensity of the C_3O lines. Fumaroyl dichloride turned out to be superior to the Meldrum's acid derivative (5,5'-bis(2,2-dimethyl-4,6-dioxo-1,3-dioxanylidene)) used in the previous work¹ due to the absence of interfering acetone lines and the higher vapor pressures obtainable by the liquid precursor. Both precursors yielded C_3O lines of comparable intensity at the same pressure.

Results

Rotational transitions of the isotopic species of tricarbon monoxide were predicted from rotational constants based on a geometry obtained by an ab initio MO calculation.² All low J transitions could be observed within 10 MHz from the predicted frequency and could be assigned by observing their Stark effect. Higher J transitions could then easily be found and assigned. The measured transition frequencies of the parent species and of all four singly substituted isotopes are listed in Table I together with the derived rotational and centrifugal distortion constants.

Unlabeled fumaroyl dichloride was used to search for vibrationally excited states of C_3O . For each ground-state line with $J > 1$ a pair of lines with about the same intensity has been found. They have been assigned as the l -type doublet of the first excited state of the lowest bending vibration. In Table II the measured transition frequencies and the derived rotational, l -type doubling and centrifugal distortion constant B_v , q_v , and D_v , respectively, are given. No other vibrational satellites of C_3O could be found.

Discussion

The discovery of the l -doubling lines of a vibrationally excited bending state is a strong argument in favor of the molecule being

(3) Kyrides, L. P. "Organic Syntheses"; J. Wiley & Sons: New York, 1955; Collect. Vol. III, pp 422–424.

(4) Murray, A.; Williams, D. L. "Organic Syntheses with Isotopes"; Interscience: New York, 1958; Vol. I, pp 129–130. Ott, D. G. "Syntheses with Stable Isotopes"; Wiley-Interscience: New York, 1981; pp 63–64.

(5) Weast, R. C., Ed. "Handbook of Chemistry and Physics"; CRC Press: Cleveland.

(6) Nystrom, R. F.; Loo, Y. H.; Leak, J. C. *J. Am. Chem. Soc.* **1952**, *74*, 3434–3435.

(7) Snyder, H. R.; Kruse, C. W. *J. Am. Chem. Soc.* **1958**, *80*, 1942–1944.

Table I. Microwave Transition Frequencies and Spectroscopic Constants of Isotopomers of C₃O^a

C ₃ O		C=C=C= ¹⁸ O		C=C= ¹³ C=O		C= ¹³ C=C=O		¹³ C=C=C=O	
transition	frequency, MHz	transition	frequency, MHz	transition	frequency, MHz	transition	frequency, MHz	transition	frequency, MHz
1 ← 0	9 621.762 (38) ^b	3 ← 2	27 433.697 (21)	3 ← 2	28 787.895 (19)	3 ← 2	28 705.411 (16)	3 ← 2	27 796.960 (16)
2 ← 1	19 243.531 (21) ^b								
3 ← 2	28 865.227 (20) ^b	4 ← 3	36 578.164 (17)	4 ← 3	38 383.764 (17)	4 ← 3	38 273.804 (14)	4 ← 3	37 062.543 (14)
4 ← 3	38 486.862 (19) ^b								
5 ← 4	48 108.504 (17) ^b	8 ← 7	73 155.300 (26)	5 ← 4	47 979.599 (68)	5 ← 4	47 842.145 (45)	5 ← 4	46 328.038 (19)
6 ← 5	57 729.962 (26) ^b								
7 ← 6	67 351.334 (29)	20 ← 19	182 869.547 (75)	6 ← 5	57 575.284 (39)	6 ← 5	57 410.421 (50)	6 ← 5	55 593.802 (68)
13 ← 12	125 076.517 (16)								
18 ← 17	173 173.711 (33)								
19 ← 18	182 792.180 (47)								
<i>B</i> ₀	4 810.889 (2) MHz 0.784 (5) kHz	4 572.294 (1) MHz 0.694 (2) kHz	4 797.966 (1) kHz	4 784.246 (1) MHz 0.619 (21) kHz	4 632.840 (2) MHz 0.708 (37) kHz				

^aNumbers in parentheses behind the observed frequencies represent one tenth of the measured line width and were used to calculate weights for the least-squares fit. Numbers in parentheses behind molecular constants represents one standard deviation of the fit. ^bPreviously reported (ref 1).

Table II. Rotational Frequencies in the First Excited State of ν₅^a

<i>J'</i> - <i>J''</i>	<i>l'</i> ← <i>l''</i>	frequency, MHz
2 ← 1	1e ← 1e	19 367.345
2 ← 1	1f ← 1f	19 421.465
3 ← 2	1e ← 1e	29 050.903
3 ← 2	1f ← 1f	29 132.134
4 ← 3	1e ← 1e	38 734.412
4 ← 3	1f ← 1f	38 842.747
5 ← 4	1e ← 1e	48 417.894
5 ← 4	1f ← 1f	48 553.282

^a*B*_v = 4848.601 (2); *D*_v = 0.000 918 (39); *q*_v = 13.537 (3) MHz.

linear. Ab initio calculations even beyond the Hartree-Fock level yield the same result.² Experimental estimates of the vibrational frequency of this mode proved to be very difficult due to the problem of reliably determining intensities under flow conditions. However, Townes and Schawlow⁸ give a relation between the *l*-type doubling constant *q*_v and the vibrational frequency ω for linear molecules:

$$q_v = f(B_e^2/\omega)$$

For typical molecules *f* has a value of about 2.6. This equation proved to be correct to within 12% for all 15 molecules which have been tested by Winnewisser and Bodenseh.⁹ Applying it to tricarbon monoxide leads to a vibrational frequency of about 150 cm⁻¹, a value which is broadly consistent with the intensity measurements. Ab initio force field calculations² yield 200 and 720 cm⁻¹ for the two bending vibrations. This confirms the assignment of the *l*-doubling lines as being due to molecules in the first excited state of the lowest bending vibration ν₅.

Having determined the rotational constants of four singly substituted isotopomers, substituting each nucleus in turn, we are able to determine the substitution structure of C₃O. Kraitchman's equations¹⁰ have been used. The derived substitution coordinates are given in Table III. All nuclei are over 0.5 Å away from the center of mass. All coordinates should therefore be reliable.

(8) Townes, C. H.; Schawlow, A. L. "Microwave Spectroscopy"; McGraw-Hill Book Company; New York, 1955, 33.

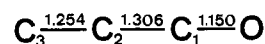
(9) Winnewisser, M.; Bodenseh, H. K. *Z. Naturforsch.* **1967**, *22A*, 1724-1737.

(10) Kraitchman, *J. Am. J. Phys.* **1953**, *21*, 17-21.

Table III. Cartesian Coordinates (Å) in the Principal Axis System of the Parent Molecule, Obtained by Kraitchman's Method^a

nucleus coordinate	C ₃	C ₂	C ₁	O
	-2.025	-0.771	0.535	1.685

^aThe atomic numbering is given in Figure 1.

**Figure 1.** The atomic numbering scheme and substitution structure (Å) of tricarbon monoxide.

Nevertheless, the center of mass condition is not fulfilled exactly. The discrepancy of 0.169 amu Å is bigger than anticipated for such a favorable case. In the isoelectronic molecule cyanoacetylene where the distances of all nuclei to the center of mass are also more than 0.5 Å, the corresponding value is 0.018 amu Å.¹¹ A possible reason for this difference is that the lowest vibration in C₃O lies lower (~150 cm⁻¹) than in cyanoacetylene (220 cm⁻¹) which therefore might have a larger influence on the ground-state rotational constant *B*₀. However, the substitution structure given in Figure 1 is the best estimate for the equilibrium structure obtainable with the present data. It can be compared with the results of an MP3/6-31G* ab initio calculation.² While the CO and the central CC bond lengths agree within 0.003 Å with the substitution structure, the terminal CC bond is calculated to be 0.017 Å larger than the experimental value. This is a fairly large discrepancy for this level of calculation. Agreements to 0.002-0.007 Å are typical for this type of calculation.

The calculated force field indicates that the lower bending vibration in C₃O is an almost pure CCC bending motion. The small terminal CC bond length could therefore result from the vibrational averaging of the rotational constant over the large-amplitude motion. A similar situation has been encountered for the CH bond length in HCNO.¹²

Acknowledgment. Dr. M. Rodler acknowledges the assistance of a Monash University Vice-Chancellor's Postdoctoral Fellowship. This work was supported by the Australian Research Grants Scheme.

(11) Costain, C. C. *J. Chem. Phys.* **1958**, *29*, 864-874.

(12) Winnewisser, B. P.; Winnewisser, M.; Winther, F. *J. Mol. Spectrosc.* **1974**, *56*, 65-94.

Registry No. I, 79864-95-2; II, 96503-55-8; III, 96503-56-9; IV, 96503-54-7; C₃O, 11127-17-6; C₃O-[¹⁸O], 11127-21-2; C₃O-[¹³C], 11127-20-1; C₃O-[²⁻¹³C], 11127-19-8; fumaroyl dichloride, 627-63-4; [¹⁸O₂]fumaroyl dichloride, 96503-53-6; [²⁻¹³C]fumaroyl dichloride,

96503-57-0; [²⁻¹³C]-(R,S)-2,3-dibromosuccinic acid, 96503-58-1; [²⁻¹³C]fumaric acid, 96503-59-2; 5,5'-bis(2,2-dimethyl-4,6-dioxo-1,3-dioxanylidene), 23340-29-6; 5-bromo-2,2-dimethyl-1,3-dioxan-4,6-dione, 66145-20-8; 5,5-dibromo-2,2-dimethyl-1,3-dioxan-4,6-dione, 66131-14-4.

On the Quencher Concentration Dependence of Fluorescence Quenching: The Role of Solution Dielectric Constant and Ionic Strength

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Abstract: A simple calculation of the dependence on quencher concentration of a fluorophore's lifetime τ is presented. Positive deviations from the linear Stern-Volmer relation are predicted and are shown to be due to the correlations among the quenchers when they are nondilute. When the fluorophores and quenchers are ions, the solution dielectric constant and ionic strength can strongly influence the magnitude of the deviations from linear Stern-Volmer behavior. For oppositely charged ions, a solution which is dilute with respect to quencher concentration can appear to be quite concentrated when the dielectric constant is low. When the intrinsic rate (the rate when the fluorophore and quencher are in contact) is not too much faster than the rate of diffusion, a regime can be reached where $1/\tau$ appears to return to linear Stern-Volmer behavior with slope given by the intrinsic rate.

The natural lifetime τ_0 of a fluorophore in solution can be shortened to τ by the presence of quenchers of concentration [Q] according to

$$\frac{1}{\tau} = \frac{1}{\tau_0} + k_f \quad (1)$$

where k_f is the contribution from quenching. For diffusion-controlled quenching by dilute quenchers $k_f = k_D[Q]$ where $k_D = 4\pi Da$ with $D = D_A + D_Q$ the sum of the fluorophore and quencher diffusion coefficients and a is their encounter separation.¹ This expression for k_f and eq 1 lead to the linear Stern-Volmer (SV) intensity law,² $I_0/I = 1 + k_D[Q]\tau_0$. If the quenchers are not dilute, the fluorophore spatial distribution about a quencher is influenced by the presence of pairs, triplets, ..., of quenchers. Therefore, k_f cannot be obtained by multiplying k_D , the result for one fluorophore and one quencher, by [Q].

In this paper we present a simple treatment of this concentration effect. We consider the case where the fluorophore and quencher may interact by a long-range potential V appropriate to ionic reactants. Then the dielectric constant and ionic strength of the solution are important determinants of the [Q] dependence of $1/\tau$ (as is true for dilute quenchers). Furthermore, we consider diffusion-influenced reactions where the intrinsic reaction rate k_0 —the rate for quenching when the fluorophore and quencher are in contact—is not necessarily much greater than k_D , as required for diffusion control.

Three regimes are identified in this work: First, when $\nu = k_0/k_D$ is small, deviations from linear SV behavior are found to be small. Second, when ν is large (diffusion control) the quencher concentration effects can be dramatic. When the reactants are of opposite charge not only is the dilute quencher rate enhanced but so is the deviation from linear SV behavior. For like charged reactants the dilute rate is reduced and so is the deviation from linear SV behavior. Third, for $\nu \gtrsim 1$ (finite ν) and high quencher concentration, the rate constant approaches the dilute reaction control result $k_f = k_0[Q]$. In this case, $1/\tau$ appears to obey a linear

SV law with slope given by the intrinsic rate constant k_0 . This last result offers the important possibility of measuring reaction rates which would ordinarily be dominated by diffusion.

Recent calculations³ of k_f for the nondilute situation have shown a faster increase than linear for k_f . In the specific context of fluorescence quenching several calculations of $1/\tau$ along with comparisons to experiment have appeared.⁴⁻⁶ Keizer⁵ has also shown how to include long-range potential effects in the nondilute case. We recently presented a systematic calculation of the concentration effect which focuses on the long-range potential case.⁷ The first correction to the dilute behavior of this calculation is exact, and we find that the simple approximate calculation presented here agrees with the systematic calculation's result.

Quenching Calculation

We introduce the calculation with the dilute, neutral (V short-ranged) case. The fluorophore number density $n(r)$ around a quencher located at the coordinate origin is, at steady state

$$n(r) = \left[1 - \frac{k_0}{k_0 + k_D} \frac{a}{r} \right] n_0 \quad (2)$$

This result is obtained by solving the diffusion equation with the radiation boundary condition⁸

$$4\pi a^2 D \left. \frac{\partial n}{\partial r} \right|_{r=a} = k_0 n(a) \quad (3)$$

and the boundary condition $n(r \rightarrow \infty) = n_0$, the bulk concentration. The radiation boundary condition states that the diffusion flux of fluorophore into the quencher equals the rate constant for

(3) Felderhof, B. U.; Deutch, J. M. *J. Chem. Phys.* **1976**, *64*, 4551. Lehenhaft, J. R.; Kapral, R. *J. Stat. Phys.* **1979**, *20*, 25. Muthukumar, M.; Cukier, R. I. *J. Stat. Phys.* **1981**, *26*, 453. Tokuyama, M.; Cukier, R. I. *J. Chem. Phys.* **1982**, *76*, 6202. Muthukumar, M. *J. Chem. Phys.* **1982**, *76*, 2667. Cukier, R. I.; Freed, K. *J. Chem. Phys.* **1983**, *78*, 2573. Keizer, J. *J. Chem. Phys.* **1983**, *79*, 4877.

(4) Peak, D.; Werner, T. C.; Denin, R. M., Jr.; Baird, J. K. *Chem. Phys.* **1983**, *79*, 3328.

(5) Keizer, J. *J. Phys. Chem.* **1982**, *86*, 5052.

(6) Keizer, J. *J. Am. Chem. Soc.* **1983**, *105*, 1494.

(7) Cukier, R. I. *J. Chem. Phys.*, in press.

(1) Schwartz, H. A.; Weston, R. E. "Chemical Kinetics"; Prentice-Hall: Englewood Cliffs, New Jersey, 1972; Chapter 6.

(2) Stern, O.; Volmer, M. *Phys. Z.* **1919**, *20*, 183.