JOURNAL OF PHYSICAL ORGANIC CHEMISTRY, VOL. 6, 367–373 (1993)

SIMULTANEOUS DETERMINATION OF PRIMARY AND SECONDARY THERMODYNAMIC ISOTOPE EFFECTS IN TAUTOMERIC EQUILIBRIA

BEN LI ZHANG, FRANÇOISE MABON AND MARYVONNE L. MARTIN*

Laboratoire de RMN et Réactivité Chimique, URA–CNRS 472, Université de Nantes, 2 rue de la Houssinière, 44072 Nantes Cedex, France

NMR determination of site-specific hydrogen isotope ratios at natural deuterium abundance (SNIF-NMR) provides the basis for simultaneous access to primary and secondary thermodynamic fractionation factors in exchange reactions and avoids the need for selective isotope labelling of the reagents. The method was applied to the measurement of fractionation parameters involving OH, CH_2, CH_3 and =CH groups in keto–enol tautomeric equilibria. The fractionation factors relating the =CH and OH sites of the enol species are simply derived from ^2H NMR spectra whereas the determination of isotope parameters which relate keto and enol positions exploits a combination of ^2H and ^1H NMR experiments. Since only monolabelled isotopomers have to be considered at natural abundance, the method also offers the advantage of avoiding the occurrence of complex equilibria associated with multi-labelled species possibly introduced by deuterium enrichment. The primary equilibrium isotope effects illustrate a preference of deuterium for the methylene fragment of the keto form with respect to the ethylenic position of the enol tautomer. Since the enol species is itself engaged in a fast tautomeric equilibrium associated with a symmetric or unsymmetric double minimum potential, the thermodynamic parameters are averaged over the exchanging partners. It is shown that the average thermodynamic fractionation factor relating the OH and =CH hydrogens of the enol are significantly influenced by the nature of the substituents at both carbonyl positions of the β-diketones. Moreover, methyl and chlorine substitution increases by a factor of about 1·1 the thermodynamic isotopic fractionation factor relating the −COCHCO− position of the keto form to the hydroxyl position of the enol.

INTRODUCTION

The thermodynamic isotope effects associated with exchange reactions at equilibrium are important sources of information for the understanding of chemical and biochemical mechanisms.[1–4] Therefore, a great deal of effort has been devoted to the determination of primary isotope fractionation factors involving exchangeable hydrogens attached to carbon or to electronegative atoms.[5–12] Secondary isotope effects associated with hydrogen situated in the vicinity of the exchanging sites have also been widely studied and the results are frequently discussed in terms of transferable substituent effects on isotope fractionation.[13,14] A number of experimental and theoretical methods have been developed for determining isotope effects on equilibrium constants. The experimental methods usually require either the previous synthesis of selectively labelled substrates or recourse to deuterium-enriched media. Under such conditions complex isotopomeric mixtures are involved in the presence of equivalent exchanging positions and the results cannot then be analysed simply in terms of fractionation factors.

The study of site-specific natural isotope fractionation by NMR (SNIF-NMR)[15,16] has been shown to provide the basis for efficient approaches to the determination of kinetic or thermodynamic fractionation parameters.[17] In particular, equilibrium liquid–vapour isotope effects associated with several isotopic species and several molecular sites can be simultaneously compared.[18] Moreover, the thermodynamic isotope effects on chemical exchanges between the hydrogens of water and of OH, SH or NH groups, for instance, are rendered directly accessible.[19]

It will be shown here that a whole set of primary and secondary thermodynamic hydrogen isotope effects can be directly determined in a one-pot experiment and without the need for selective deuterium labelling. The method was applied to the investigation of the keto–enol tautomeric system which enables fraction-

* Author for correspondence.

0894–3230/93/060367–07$08.50

Received 17 June 1992
Revised 8 February 1993

ation factors of several carbon- and oxygen-bound hydrogens to be directly compared.

EXPERIMENTAL

Materials. All the β-diketones were commercial analytical-reagent grade reagents which were used without further purification. The compounds were carefully dried since traces of water may be the source of intermolecular exchanges[20,21] which broaden the ^2H NMR signal of the hydroxyl group. In cases where the keto or enol content is very low ($K_H > 14$ or $< 0{\cdot}2$), a slight deuterium enrichment at the OH, =CH and CH_2 positions was also performed by chemical exchange with a small amount of water characterized by an isotopic ratio of about 1000 ppm. The product was then carefully dried using molecular sieves. However, only the exchanging sites can be compared in such experiments whereas a more complete set of thermodynamic factors are accessible at natural abundance.

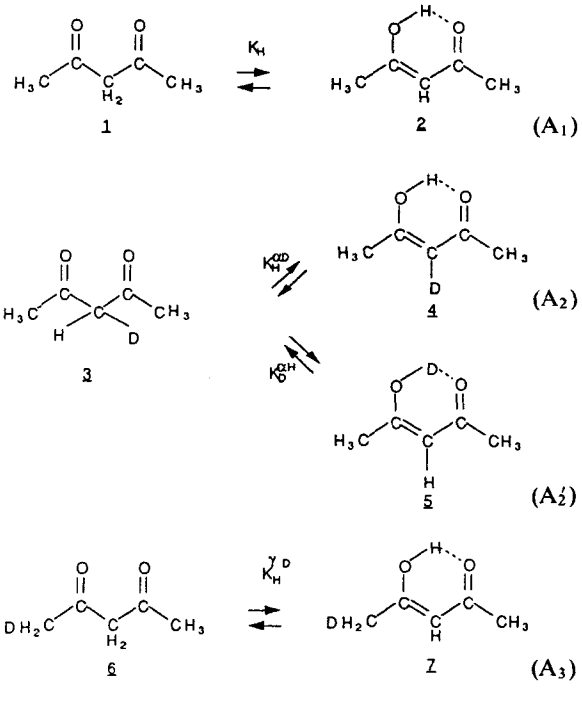
NMR experiments. The ^1H NMR spectra were recorded on Bruker WH 90 and WM 250 spectrometers. In order to avoid saturation in ^1H NMR, the pure sample was introduced into a capillary placed in a 5 mm tube which itself contained $CDCl_3$ for field-frequency locking. The delay between pulses was longer than 10 s in order to ensure complete proton relaxation; 240–500 scans were accumulated. The ^2H NMR spectra were recorded on a Bruker AM 400 spectrometer tuned to the deuterium resonance frequency (61·4 MHz) and equipped with a fluorine field-frequency locking device. C_6F_6 was used as internal locking material. The experimental requirements for quantitative ^2H NMR determinations have been discussed previously.[17] The acquisition parameters were as follows: acquisition time, 6·8 s; sweep width, 2400 Hz; active memory size, 32 K; proton broad band decoupling: exponential multiplication of the free induction decay associated with line broadening of 0·1, 0·5, 1 or 2 Hz; and number of scans, 3000–26,000. Owing to the strong prevalence of quadrupolar relaxation, nuclear Overhauser contributions are negligible. A signal-to-noise ratio higher than 100 is a prerequisite for obtaining a suitable precision of the quantitative NMR determinations. It has been checked in a collaborative experiment[22] that the reproducibility on intensity ratios measured by ^2H NMR can be better than 1% under carefully adjusted experimental conditions and for suitable samples.

The ^1H and ^2H NMR spectra were run at 303 ± 2 K. We checked that the effect of small variations of the temperature on the equilibrium constants can be neglected.

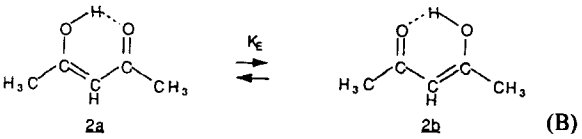
The signal areas were measured by curve-fitting procedures (NMR 1 software, New Methods, Syracuse, NY, USA). The assignments of the deuterium signals were derived from straightforward analysis of the proton spectra.

PRINCIPLES OF THE METHOD

At the low natural abundance of deuterium (*ca* 150×10^{-6}) only monodeuterated isotopomers have to be considered. In the case of acetylacetone, for instance, two fully protonated species **1** and **2** and five monolabelled species **3**–**7** are engaged in the keto–enol equilibria A_1–A_3



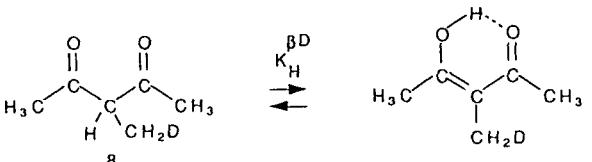
Moreover the enol species **2**, **4**, **5** and **7** undergo hydrogen exchanges of type B.



Whereas the tautomeric equilibria (A) are slow on the NMR time scale, the enolic exchanges (B) are fast and the ^1H and ^2H NMR parameters of the enol are means over those of the individual species such as **2a** and **2b**. Consequently, only one signal is observed for both methyl groups of **2**, **4**, **5** or **7**.

No intramolecular competition, such as between reactions (A_2) and (A_2') occurs when the keto form is substituted in position 3 and therefore no secondary α

isotope effect may intervene. However, if the substituent is an alkyl group a secondary β equilibrium isotope effect is possibly involved since the equilibrium constant $K_H^{\beta D}$ of reaction (A_4) may differ from the equilibrium constant K_H associated with the corresponding fully protonated species.



8 9

(A_4)

Equilibria (A_1) on the one hand and (A_2)–(A_4) on the other can be investigated in the same medium by a combination of ^2H and ^1H NMR experiments.

Thermodynamic fractionation factors, ϕ compare the relative affinity of the hydrogen isotopes for a given molecular site, i, of compound A to that for another site j pertaining either to the same molecule, A, or to another molecule, B. The relative fractionation parameter $\phi_{A_i}^{B_j}$ is defined as

$$\phi_{A_i}^{B_j} = (D/H)_{A_i}/(D/H)_{B_j} \qquad (1)$$

where (D/H) denotes the ratio of the numbers of deuterium and protium atoms in position i of A and j of B (or A). This equation can be rearranged into

$$\phi_{A_i}^{B_j} \approx \frac{D_{A_i}}{D_{B_j}} \times \frac{P_{B_j} N_B}{P_{A_i} N_A} \qquad (2)$$

where P_{A_i} and P_{B_j} are the stochiometric numbers of hydrogens at sites A_i and B_j and N_A and N_B are the numbers of moles of compounds A and B. The ratio of the numbers of deuterium atoms in sites A_i and B_j, D_{A_i}/D_{B_j}, is directly accessible from the ^2H NMR spectrum.

Water is frequently used as a reference for characterizing isotope effects. The isotope fractionation factor of site A_i with respect to water, $\phi_{A_i}^{W}$, can be measured directly or calculated from $\phi_{A_i}^{B_j}$ and from the fractionation factor of B_j with respect to water:

$$\phi_{A_i}^{W} = \phi_{A_i}^{B_j} \phi_{B_j}^{W} \qquad (3)$$

In the very dilute isotopic conditions of natural abundance only monolabelled isotopomers have to be taken into account. Therefore, the method avoids the need for separating the roles of differently labelled fragments, such as $-CHD-$ and $-CD_2-$, which are present in enriched systems. In particular, in exchanges involving aqueous media at natural isotopic abundance, the results are independent of the isotopic equilibrium constant of water, K_W, which is known to differ significantly from the statistical value of 4:

$$H_2O + D_2O \overset{K_W}{\rightleftharpoons} 2HOD$$

The ^2H NMR method is particularly suited to comparing the relative affinities of the hydrogen isotopes for the exchanging $=CH$ and OH positions in the enol structure (e). The fractionation parameter $\phi_{CH(e)}^{OH(e)}$ is then easily accessible from the areas, S, of the $=CD$ and OD signals in the deuterium spectrum

$$\phi_{CH(e)}^{OH(e)} = \frac{(D/H)_{CH(e)}}{(D/H)_{OH(e)}} = \frac{S_{CD}^{(e)}}{S_{OD}^{(e)}} \qquad (4)$$

Similarly, the primary fractionation parameter $\phi_{CH_2(k)}^{OH(e)}$ which compares the relative hydrogen isotope affinity for the $-COC(H)CO-$ fragment in the keto (k) form with that for the enol (e) hydroxyl position, is directly related to the thermodynamic constants of equilibria (A_1) and (A_2'):

$$\phi_{CH_2(k)}^{OH(e)} = \frac{1}{2} \times \frac{K_H}{K_D^{\alpha H}} = \frac{1}{2} \times \frac{S_{CD(H)}^{(k)}}{S_{OD}^{(e)}} \times \frac{a}{1-a} \qquad (5)$$

where $S_{CD(H)}^{(k)}$ and $S_{OD}^{(e)}$ are the areas of the keto $-CD(H)-$ and enol OD signals in the deuterium spectrum and a is the equilibrium mole fraction of the enol form, which can be derived from the ^1H spectrum. It should be noted that the coefficient 1/2 disappears when the methylene position of the keto tautomer is substituted.

Another fractionation factor may refer the $-COCH_2CO-$ fragment of the keto tautomer to the ethylenic position in the enol [equilibrium (A_2)]:

$$\phi_{CH_2(k)}^{CH(e)} = \frac{1}{2} \times \frac{K_H}{K_H^{\alpha D}} = \frac{1}{2} \times \frac{S_{CDH}^{(k)}}{S_{CD}^{(e)}} \times \frac{a}{1-a} \qquad (6)$$

The secondary thermodynamic isotope parameters characterizing partner sites not directly involved in the exchange process are defined in the same way. Thus the relative affinity of CH_3 and CH_2D groups towards the ketone, $-COCH_2$, and enol, $-C(OH)=CH$, moieties is expressed as [equilibrium(A_3)]

$$\phi_{CH_3(k)}^{CH_3(e)} = \frac{K_H}{K_H^{\gamma D}} = \frac{S_{CH_2D}^{(k)}}{S_{CH_2D}^{(e)}} \times \frac{a}{1-a} \qquad (7)$$

S_{CH_2D} being the area of the methyl signals of the enol and ketone tautomers in the ^2H NMR spectrum.

It should be emphasized that the present method offers the unique capability of simultaneously comparing the thermodynamic factors associated with all diastereotopic positions in the partners of a given equilibrium [equations (4)–(7)]. These isotopic effects can be directly obtained with an accuracy which is in fact that of quantitative NMR determinations performed in relatively favourable conditions for small organic molecules, since high signal-to-noise ratios are rapidly obtained (1–3 h) for concentrated solutions (short longitudinal relaxation times) and discriminating

nuclear Overhauser effects may usually be neglected for the quadrupolar deuterium nucleus.

RESULTS AND DISCUSSION

In spite of more than 20 years of continuing interest, the number of experimental determinations of thermodynamic isotope fractionation factors remains small.[13,23] Moreover, scattered values are often published for a given equilibrium. This situation is probably due to the limited number of exchanging systems accessible to the existing techniques and to the difficulties in implementing reliable procedures. Tautomeric equilibria are well suited to a ^2H NMR investigation of site-specific isotope fractionation at equilibrium. As discussed in the previous section, it is possible, in principle, to determine in a single experiment isotope fractionation parameters which refer the relative deuterium–protium affinity for different sites in the keto form to that for associated positions in the enol tautomers. Moreover, the method provides simultaneous access to the thermodynamic fractionation factor which relates the hydroxyl and ethylenic positions in the enol structure. It should also be emphasized that at natural isotopic abundance, the risk is avoided of pertubations in deuterium contents which may occur in enriched media owing to exchange with atmospheric water.

The values of the equilibrium constants and fractionation parameters obtained in the investigation of tautomeric equilibria involving differently substituted β-diketones are given in Table 1.

Owing to the sensitivity of the tautomeric equilibria to medium and temperature effects,[20] literature values[24] of the thermodynamic constant, K_H, associated with the keto–enol equilibrium of the fully protonated species cannot be safely used for the determination of the isotope fractionation factors. Consequently, K_H was measured by ^1H NMR on the investigated samples.

In principle, the experimental conditions for the determination of the 'intramolecular' enol fractionation factor $\phi^{OH(e)}_{CH(e)}$ are specially favourable since the signals to be compared are obtained in a single spectrum and exhibit similar intensities [equation (4)]. However the accuracy of the area determinations may be degraded in certain cases as a result of line broadening of the hydroxyl signal. In the case of keto–enol comparisons [equations (5)–(7)] errors occurring in both ^1H and ^2H quantitative determinations are cumulated. The repeatability and reproducibility of the method were estimated by performing, for a given β-diketone, a number *NE* of independent series of experiments, each series comprising a number *NR* of successive experiments. These series of deuterium and proton spectra were run at different periods using different sample tubes. The results are detailed in Table 1 for compound **2**. The relative standard deviation of the repeatability of the NMR determinations is usually of the order of 2% and that of the reproducibility (long-term replications) is slightly less than 2·5%. However, it should be emphasized that the accuracy of area determinations is strongly dependent on the signal-to-noise ratio and on the shape of the signals. Less favourable conditions are encountered in the presence of broad hydroxylic signals and for high or low values of K_H. In the latter case, possible deviations in the isotope contents with respect to the mean statistical value mainly affect the small signals of the minor species, which are more exposed to sensitivity limitations. Although a reasonable repeatability is reached in a given series of experiments, the results corresponding to very diluted enols (Table 1) are more likely to be affected by systematic errors.

The measured keto–enol fractionation factors relate specific hydrogen sites in the keto form to positions situated in less well defined environments in the enol. Indeed, various investigations of the structure of the hydrogen bridge in the enol of acetylacetone and of isotope effects in keto-enol equilibria have given rise to some controversial results and interpretations.[20,21,25–28] It is now recognized that the hydrogen-bonded proton of the enol is not located in a single potential well but is rapidly transferred between the minima of a symmetric double potential well.[21] When the R_1 and R_3 substituents of the β-diketones, $R_1COCH(R_2)COR_3$, are different the enol species evolves in an unsymmetric double minimum potential. The fractionation factors (Table 1) then characterize environments of the enol positions which correspond to weighted averages over the exchanging partners in equilibria of type B. In the case of the keto esters the situation is simpler since the enol structures are present nearly exclusively in the enol tautomeric form with the hydroxyl opposite to the ester side.[29] In this respect, it should be noted that information on the preferential direction of enolization can be obtained by measuring isotope effects on the ^{13}C chemical shifts of the enol species.[30,31]

The behaviour of the fractionation factor, $\phi^{OH(e)}_{CH(e)}$ (Table 1), shows that the relative affinity of the hydrogen isotopes for the hydroxyl site and for the carbon-bound ethylenic position is significantly influenced by a change in the remote R_1 and R_3 substituents. Similarly, the secondary thermodynamic isotope effect associated with deuteurium substitution in the R_1 methyl group may be significantly different from unity. As illustrated by the values of $\phi^{CH(e)}_{CH_2(k)}$, it is also observed that the heavy atom exhibits a higher preference for the sp^3-hybridized carbon of the keto form than for the sp^2-hybridized carbon of the enol.

From a theoretical point of view, the thermodynamic fractionation factors can be calculated from the ratio of the reduced partition functions, *Q*, of the exchanging

Table 1. Thermodynamic isotope constants associated with keto–enol equilibria of type (A_1) to (A_4) (see text) involving β-diketones $R_1COCHR_2COR_3$.[a]

Compound	R_1	R_2	R_3	$NR \times NE$	K_H	$NR \times NE$	$K_H^{\alpha D}$	$K_D^{\alpha H}$	$K_H^{\gamma D}$	$K_H^{\beta D}$	$\phi_{CH(e)}^{OH(e)}$	$\phi_{CH_2(k)}^{CH(e)}$	$\phi_{CH_2(k)}^{OH(e)}$	$\phi_{\gamma CH_3(k)}^{\gamma CH_3(e)}$	$\phi_{\beta CH_3(k)}^{\beta CH_3(e)}$
1	CH_3	H	CH_3	5 × 3	4·42 (0·05)	4 × 3	2·17 (0·03)	2·03 (0·07)	4·20 (0·07)		1·07 (0·04)	1·02	1·09	1·05	
				6 × 1	0·57 (0·02)	4 × 1		0·50 (0·01)	0·66 (0·01)	0·59 (0·01)			1·14	0·86	0·97
				5 × 1	0·58 (0·02)	5 × 1		0·52 (0·02)	0·66 (0·01)	0·59 (0·01)			1·12	0·88	0·98
2	CH_3	CH_3	CH_3	5 × 1	0·59 (0·02)	3 × 1		0·53 (0·02)	0·66 (0·02)	0·60 (0·02)			1·11	0·89	0·98
				Mean	0·58 (0·02)			0·52 (0·02)	0·66 (0·02)	0·59 (0·02)			1·12	0·88	0·98
3	CH_3	Cl	CH_3	6 × 1	14·4 (0·1)	4 × 1		14·77 (0·47)						0·97	
4	CH_3	H	OC_2H_5	6 × 1	0·079 (0·001)	5 × 1	0·038 (0·001)	0·045 (0·001)			0·83 (0·01)	1·04	0·88		
5	CH_3	H	$O(CH_3)_3$	6 × 1	0·087 (0·002)	5 × 1	0·044 (0·001)	0·052 (0·001)			0·84 (0·03)	0·99	0·84		
6	CH_2Cl	H	OC_2H_5	4 × 1	0·098 (0·002)	5 × 2	0·046 (0·001)	0·047 (0·001)			1·00 (0·02)	1·06	1·04		
7	CH_3	Cl	OC_2H_5	6 × 1	0·169 (0·002)	6 × 1		0·149 (0·003)					1·13		
8	CF_3	H	OC_2H_5	4 × 2	6·99 (0·06)	5 × 2	2·93 (0·08)	3·31 (0·10)			0·89 (0·01)	1·19	1·06		
9	CF_3	H	CF_3		[b]						0·92 (0·02)				

[a] The isotopic fractionation factors, ϕ, relate specified positions of the keto (k) and enol (e) structures [equations (5)–(7)] or the two exchanging sites, =CH and OH, of the enol tautomer [equation (4)]. Each determination of the isotopic equilibrium constants is obtained from *NE* independent series of *NR* successive experiments. The standard deviations characterizing the repeatability are given in parentheses.

[b] No keto form has been observed.

isotopic partners:[2,5,32]

$$A(H) + B(D) \overset{K_A^B}{\rightleftharpoons} A(D) + B(H)$$

$$\phi_A^B = \frac{(Q_{AD}/Q_{AH})_{red}}{(Q_{BD}/Q_{BH})_{red}} = \frac{(s_D/s_H)_B}{(s_D/s_H)_A} \times K_A^B \qquad (8)$$

where $(Q_{AD}/Q_{AH})_{red}$ and $(Q_{BD}/Q_{BH})_{red}$ are the ratios of the partition functions divided by the corresponding ratios calculated classically and $(s_D/s_H)_{A,B}$ are the ratios of the symmetry numbers associated with the A and B isotopic species. The reduced partition function ratios can be expressed as a function of the vibrational frequencies of the molecules, either determined experimentally or derived from quantum-mechanical force constants computed with different levels of approximation.[5,33–35] Detailed analyses of the vibrational frequencies of the β-diketones would be necessary to interpret the fractionation factors quantitatively. However, it is observed that the direct isotope effects illustrated for instance by the $\phi^{CH(e)}_{CH_2(k)}$ parameter are consistent with a stronger lowering of the vibrational frequencies associated with the ketone structure, as compared with the enol ethylenic moiety, under the influence of deuterium substitution.[36,37] This behaviour is in qualitative agreement with the existence of higher vibrational frequencies in the ketone as compared with the enol structure and in particular with the occurrence of low out-of-plane bending frequencies in the enol.[38] Moreover, since the heavier atom exhibits an increasing preference for positions where it is bound more stiffly,[1] i.e. for positions which correspond to higher zero point energy, the fractionation factors involving the enol hydroxyl, $\phi^{OH(e)}_{CH(e)}$ and $\phi^{OH(e)}_{CH_2(k)}$, are also expected to vary with the strength of the intramolecular hydrogen bond.

In another approach, empirical or theoretical substituent scales have been derived which enable, for example, the fractionation factor of a hydrogen on an sp^3-hybridized carbon atom in a CHXYZ environment to be calculated from the fractionation factor of CH_4 and isotopic substituent effects, δ:[7,13,14,35,39]

$$\phi^W_{CHXYZ} = \phi^W_{CH_4}\, \delta_X\, \delta_Y\, \delta_Z \qquad (9)$$

Several scales of isotopic substituent parameters have been proposed.[13,14,35] In this approach, it is generally considered that isotopic fractionation effects are mainly conditioned by the nature of the atom directly bound to the carbon. It has been shown that the δ parameter is higher than unity for a methyl substitution and increases with increasing electronegativity of the substituent. The results given in Table 1 are in satisfactory agreement with these latter trends. In particular, the increase in the $\phi^{OH(e)}_{CH_2(k)}$ factor on going from acetylacetone (**1**) to the diketone **2** substituted in position 3 by a methyl group is consistent with a substituent effect of the order of 1·1.[35] Similarly, the replacement of H by Cl on C-2 of the ethyl 3-oxobutanoate **4** is accompanied by an increase in $\phi^{OH(e)}_{CH(k)}$ corresponding to a substituent effect of 1·16. However, the data also illustrate a considerable sensitivity to remote structural differences.

REFERENCES

1. J. Bigeleisen, *Science* **147**, 463 (1965).
2. M. Wolfsberg, *Acc. Chem. Res.* **5**, 225 (1972).
3. P. F. Cook, J. S. Blanchard and W. W. Cleland, *Biochemistry* **19**, 4853 (1980).
4. W. W. Cleland, *Methods Enzymol.* **64**, 104 (1980); *Bioorg. Chem.* **15**, 283 (1987).
5. S. R. Hartshorn and V. J. Shiner, *J. Am. Chem. Soc.* **94**, 9002 (1972).
6. M. M. Kreevoy, R. Eliason, R. A. Landholm, T. S. Strauss and J. L. Melquist, *J. Phys. Chem.* **76**, 2951 (1972).
7. H. P. Meloche, C. T. Monti and W. W. Cleland, *Biochim. Biophys. Acta* **480**, 517 (1977).
8. J. H. Rolston and K. L. Gale, *J. Phys. Chem.* **88**, 4394 (1984).
9. M. Saunders, S. Saunders and C. A. Johnson, *J. Am. Chem. Soc.* **106**, 3098 (1984).
10. R. Bone and R. Wolfenden, *J. Am. Chem. Soc.* **107**, 4772 (1985).
11. R. M. Jarret and M. Saunders, *J. Am. Chem. Soc.* **107**, 2648 (1985).
12. J. Reuben, *J. Am. Chem. Soc.* **108**, 1082 (1986).
13. A. J. Kresge, R. A. More O'Ferrall and M. F. Powell, in *Isotopes in Organic Chemistry*, edited by E. Buncel and C. C. Lee, Vol. 7, p. 177. Elsevier, Amsterdam (1987).
14. P. Scharlin, *Acta Chem. Scand.* **43**, 244 (1989).
15. G. J. Martin and M. L. Martin, *Tetrahedron Lett.* **22**, 3525 (1981).
16. G. J. Martin, M. L. Martin, F. Mabon and M. J. Michon, *J. Chem. Soc., Chem. Commun.* 616 (1982).
17. M. L. Martin and G. J. Martin, in *NMR Basic Principles and Progress*, edited by P. Diehl, E. Fluck, H. Günther, R. Kosfeld and J. Seeling, Vol. 23, p. 1. Springer, Berlin (1991).
18. I. Moussa, N. Naulet, M. L. Martin and G. J. Martin, *J. Phys. Chem.* **94**, 8303 (1990).
19. B. L. Zhang and M. L. Martin, *J. Am. Chem. Soc.* **114**, 7089 (1992).
20. M. J. T. Robinson, K. M. Rosen and J. D. B. Workman, *Tetrahedron* **33**, 1655 (1977).
21. W. Egan, G. Gunnarsson, T. E. Bul and S. Forsen, *J. Am. Chem. Soc.* **99**, 4568 (1977).
22. C. Guillou, M. Trierweiler and G. J. Martin, *Magn. Res. Chem.* **26**, 491 (1988).
23. W. A. Van Hook, *J Phys. Chem.* **76**, 3040 (1972).
24. J. L. Burdett and M. T. Rogers, *J. Am. Chem. Soc.* **86**, 2105 (1964).
25. A. H. Lowrey, C. George, P. D'Antonio and J. Karle, *J. Am. Chem. Soc.* **93**, 6339 (1971).
26. D. W. Thompson and A. L. Allred, *J. Phys. Chem.* **75**, 433 (1971).
27. D. B. Dahlberg and F. A. Long, *J. Am. Chem. Soc.*. **95**, 3825 (1973).
28. L. J. Altman, D. Laungani, G. Gunnarsson, H. Wennerström and S. Forsen, *J. Am. Chem. Soc.* **100**, 8264 (1978).

29. C. G. C. Geraldes, M. T. Barros, C. D. Maycock and M. I. Silva, *J. Mol. Struct.* **238**, 335 (1990).
30. P. E. Hansen, *Magn. Reson. Chem.* **24**, 903 (1986).
31. C. Engdahl, A. Gogoll and V. Edlund, *Magn. Reson. Chem.* **29**, 54 (1991).
32. J. Bigeleisen and M. G. Mayer, *J. Chem. Phys.* **15**, 261 (1947).
33. D. Z. Goodson, S. K. Sarpal, P. Bopp and M. Wolfsberg, *J. Phys. Chem.* **86**, 659 (1982).
34. S. K. Sarpal, *J. Chem. Soc., Faraday Trans.* **85**, 1921 (1989).
35. I. H. Williams, *J. Phys. Org. Chem.* **3**, 181 (1990).
36. A. Streitwieser, R. H. Jagow, R. C. Fahey and S. Suzuki, *J. Am. Chem. Soc.* **80**, 2326 (1958).
37. J. M. Jones and M. L. Bender, *J. Am. Chem. Soc.* **82**, 6322 (1960).
38. B. Wojtkowiak and M. Chabanel, *Spectrochimie Moléculaire*, pp. 205–209. Technique et Documentation, Paris (1977).
39. M. I. Schimerlik, J. E. Rife and W. W. Cleland, *Biochemistry* **14**, 5347 (1975).