$BuMe_2SiCl$;[3a,b,e] (C) treatment of a ketone or aldehyde in THF with KH followed by Me_3SiCl or *t*-$BuMe_2SiCl$;[4c,d,f] (D) treatment of THF with *n*-BuLi to generate acetaldehyde enolate followed by *t*-$BuMe_2SiCl$.[3c] The silyl ethers were purified by gas chromatography and their identities and purities confirmed by NMR examination. The *E*/*Z* isomeric pairs **15**/**16** and **17**/**18** were separated by VPC by using a 6 mm × 3 m TCEP on Chromosorb P column. Differentiations of **15**/**16**[3b] and **17**/**18**[4b,c] were confirmed by comparison with their published NMR spectra. With the exception of **12** and **13**, trimethylsilyl ethers corresponding to each of the structural types **1**–**19** have been reported in the literature. Correct elemental analyses (C, H) and spectral data were obtained for **12** and **13**. Methods of preparation (A–D above), wavelengths (nm) for kinetics studies, and previous literature references were as follows: **1** (A) 215;[3c,d] **2** (A) 243;[4a] **3** (A) 228;[4e] **4** (B) 230;[3e] **5** (D) 215;[3c] **6** (C) 262;[4f] **7** (C) 228; **8** (C) 225; **9** (A) 280;[4a] **10** (A) 225;[4a] **11** (A) 223;[4a] **12** (A) 225; **13** (A) 265; **14** (A) 228;[3f] **15** (A) 261;[3b] **16** (A) 256;[3b] **17** (A) 257;[4b,c] **18** (A) 257;[4b,c] **19** (A) 272.[9]

Kinetics were measured by observing the change of the alkene or carbonyl absorption in the UV by using Cary 14 or 118 spectrophotometers. Buffer solutions were prepared by mixing aliquots of aqueous HA, NaOH, and NaCl solutions to a total of 50 mL (ionic strength $\mu = 0.10$) and diluting to 100 mL with spectral-grade acetonitrile (Matheson Coleman and Bell). Proton concentrations of the buffer solutions were measured with Beckman Zeromatic or Corning 130 pH meters. Acid concentrations of the HCl solutions were measured by titration. For the kinetic runs 3 mL of catalyst solution was equilibrated in 1-cm cells in the thermostated cell compartments and the alkene was added either neat or dissolved in acetonitrile to give solutions approximately 2×10^{-3} M in alkene. The solutions were shaken and the change of the UV absorption was monitored with time.

Data for $Me_3SiOCMe{=}CH_2$ illustrating the formate buffer catalysis at different buffer ratios and the dependence of k_{cat} (the catalytic constant for the buffer-catalyzed reaction) on the mole fraction HCO_2H of the total buffer species present are shown in Figure 3.

Acknowledgment is made to the Natural Science and Engineering Research Council of Canada and the Undergraduate Research Fund of Scarborough College for support of this work.

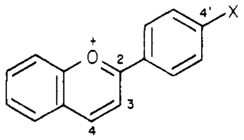
Hydration of the Flavylium Ion

Robert A. McClelland* and Sherrin Gedge

Contribution from the Department of Chemistry, University of Toronto, Scarborough College, West Hill, Ontario, Canada M1C 1A4. *Received February 15, 1980*

Abstract: A spectral and kinetic investigation has been carried out of the transformations undergone in aqueous solution by the parent flavylium ion and its 4′-methyl and 4′-methoxy derivatives. Evidence is reported for the existence at some time under some condition of seven species, the flavylium ion (F^+), two pseudobases, a 2-hydroxy adduct (B2) and a 4-hydroxy adduct (B4), the *cis*-2-hydroxychalcone and its ionized form (cC and cC^-), and the *trans*-2-hydroxychalcone and its ionized form (tC and tC^-). At pH 6.5–8.5, F^+ is relatively rapidly hydrated producing a mixture of B4, B2, and cC. The latter two are in equilibrium, their equilibration proceeding far more rapidly than F^+ is hydrated. B4 is a kinetic product of the hydration only; over a short period of time it rearranges via F^+ to the equilibrium mixture of B2 and cC. In base solution the behavior is similar, but the B2 $\rightleftharpoons$ cC equilibrium is displaced toward the chalcone since it ionizes. Thus, at pH 12, F^+ reacts very rapidly with OH^- producing a mixture of B4 and cC^-, the latter coming from B2 initially formed. This is followed by the rearrangement of B4, the kinetic product, to cC^-. In acid solutions, pH 2–6, an acid–base type of equilibrium is relatively rapidly established between the cationic F^+ and the neutral species B2 and cC. In all of these solutions, with the exception of strong acids, a slow further reaction occurs resulting eventually in complete transformation to tC or, in base, tC^-. The upper limit on the relative amount of *cis*-chalcone or pseudobase present in equilibrium with the *trans*-chalcone after complete reaction is 0.02%. For a scheme $B4 \rightleftarrows F^+ \rightleftarrows B2 \rightleftarrows cC\ (\rightleftarrows cC^-) \rightarrow tC\ (\rightleftarrows tC^-)$, rate constants and equilibrium constants for each reaction stage have been obtained by a kinetic and spectral analysis.

The anthocyanin pigments responsible for a variety of plant and flower colors have been known for some time to be derivatives of polyhydroxyflavylium ions.[1-3] Also well established[4,5] is the fact that these colored cations are stable only in relatively acidic solutions, undergoing various structural transformations when placed in less acidic, neutral, or basic media, often with quite dramatic color changes or color disappearance. This chemistry has now been examined for a number of these cations,[4,5] including various anthocyanins, anthocyanidins, and synthetic flavylium salts.[6] There appears, however, not to have been a detailed investigation of the parent ion. We report here such a study, for three ions (F^+), the parent and two 4′-substituted derivatives.



F^+, X = H, Me, MeO

Results

Spectra of Flavylium Ions, pH 1–7, and pK_a Determination. Our general experimental approach was to prepare a stock solution in 0.1 M HCl of the perchlorate salt of each flavylium ion and to adjust this to the desired pH by mixing with appropriate buffers. The flavylium ions appear to be almost indefinitely stable in these stock solutions, in that there is little change over a period of days or even weeks in the absorption spectra of the solutions. One such spectrum showing the characteristic visible absorption peak of the flavylium ion is drawn in Figure 1.

Adjustment of the pH of the stock solution to 7 results in rapid decolorization, a spectrum taken as quickly as possible showing several overlapping peaks below 300 nm, but having little residual absorbance in the visible region. One example of such a spectrum can be seen in the curve labeled neutral-initial in Figure 1.

---

(1) Willstätter, R.; Everest, A. E. *Justus Liebigs Ann. Chem.* **1913**, *401*, 1913.

(2) Pratt, D. D.; Robinson, R. *J. Chem. Soc.* **1922**, 1577.

(3) Hayashi, K. In "The Chemistry of Flavonoid Compounds", Geissman, T. A., Ed.; Pergamon Press: Oxford, 1962; pp 248–285.

(4) Jurd, L. In "The Chemistry of Plant Pigments", Chichester, C. O., Ed.; Academic Press: New York, 1972; p 123.

(5) Jurd, L. *Recent Adv. Phytochem.* **1972**, 135.

(6) Anthocyanidins have a 3-hydroxy substituent, and in anthocyanins this group is glycosylated. In the synthetic flavylium salts studied this position has been unsubstituted.

0002-7863/80/1502-5838$01.00/0

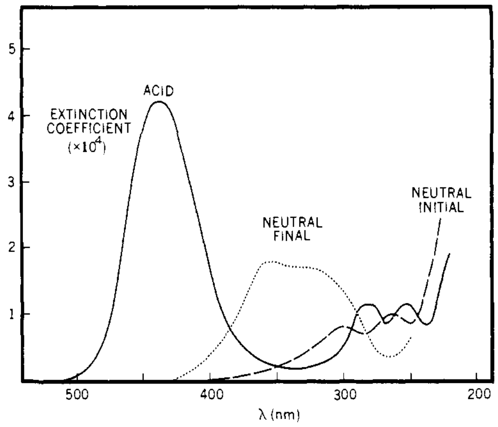


Figure 1. Spectra of 4′-methoxyflavylium perchlorate in 0.1 M HCl (—) and dilute phosphate buffer, pH 7, the curve (– –) being obtained within 20–60 s of neutralization of the stock acid solution, and the curve (-----) after 6 days. The latter curve is also obtained for a solution of *trans*-2-hydroxy-4′-methoxychalcone in the pH 7 buffer.

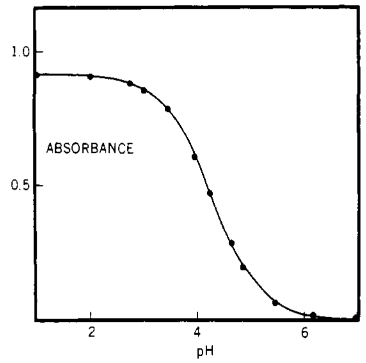


Figure 2. Absorbance at 439 nm of 2.1×10^{-5} M solutions of 4′-methoxyflavylium perchlorate, obtained 60 s after neutralization or dilution of stock acid solution. The curve drawn is the titration curve for an acid with $pK^a_{obsd} = 4.29$ and limiting absorbances of 0.915 in acid and 0.0 in base.

Whatever has occurred on neutralization is readily reversible. Immediate reacidification of the neutralized solution results in a rapid and quantitative return of the flavylium ion spectrum.

In solutions of pH intermediate between 1 and 7, a set of spectra result which show the characteristics of a simple acid–base equilibrium:

$$F^+ \rightleftharpoons \text{neutral} + H^+ \qquad (1)$$

with the equilibrium favoring F^+ at pH 1 and the "neutral" form at pH 7. This equilibrium is uncomplicated by further reactions or side reactions at least over a short period of time as revealed by the presence of good isosbestic points as the spectra change as a function of pH. Numerical values of acidity constants appropriate to eq 1 were evaluated from the change in absorbance at the λ_{max} of the flavylium ion (see Figure 2), according to the equation

$$K^a_{obsd}(F^+) = \frac{[\text{neutral}][H^+]}{[F^+]} = \frac{A_{acid} - A}{A - A_{neut}}[H^+] \qquad (2)$$

where A_{acid} and A_{neut} refer to acid and neutral solutions and A is the absorbance at intermediate pH. (The reader should note that, to distinguish between observed and derived parameters, we will define empirical equilibrium constants and rate constants with a subscript "obsd", as in eq 2. The derived parameters will be defined later. Values of empirical constants are to be found in Table I.)

Acidity constants have now been reported for a number of flavylium ions,[7-15] with the "neutral" species of eq 1 having various structures: an anhydro base,[13-15] a pseudobase,[7,9-15] or a chalcone.[8,16] We will defer discussion of the identity of the "neutral" form in our systems until later.

Table I. Observed Quantities (25 °C, Ionic Strength = 0.1)

quantity	4'-MeO	4'-Me	unsubst
$pK^a_{obsd}(F^+)$ (2)[a]	4.29	3.59	2.98
$pK^a_{obsd}(\text{base})$ (3)	9.43 (spect) 9.48 (kinet)		10.24 (kinet)
$k^{base}_{obsd}(tC)$, s^{-1} (3)	4.6×10^{-5}		2.6×10^{-4}
$k^{neut}_{obsd}(tC)$, s^{-1} (3)	1.95×10^{-5}		2.5×10^{-5}
$k^{W}_{obsd}(\text{dis})$, s^{-1} (5)	1.38	5.9	12.7
$k^{OH}_{obsd}(\text{dis})$, $M^{-1}\,s^{-1}$ (5)	1.8×10^5	5.2×10^5	7.2×10^5
$k^{H}_{obsd}(\text{isom})$, $M^{-1}\,s^{-1}$ (6)	9.0×10^5	4.4×10^5	2.5×10^5
$k^{W}_{obsd}(\text{isom})$, s^{-1} (6)	0.0015		
$k^{H}_{obsd}(F^+\text{-fast})$, $M^{-1}\,s^{-1}$ (8)	1.38×10^4	8.4×10^3	4.7×10^3
$k^{H}_{obsd}(F^+\text{-slow})$, $M^{-1}\,s^{-1}$ (7)	39	44	42
$k^{W}_{obsd}(F^+\text{-slow})$, s^{-1} (7)	0.46	0.96	1.78
$\Delta OD(\text{slow})/\Delta OD(\text{fast})$	0.50	0.16	0.06
$k^{OH}_{obsd}(B2 \rightleftharpoons cC)$, $M^{-1}\,s^{-1}$	2×10^{10}	6×10^{10}	1×10^{11}

[a] The number in parentheses refers to the equation in the text defining the constant.

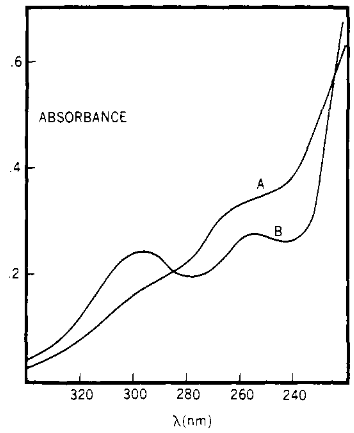


Figure 3. Spectra of 4′-methoxyflavylium ion in Tris buffer, pH 8.3. Curve A was obtained 20–40 s after mixing of the stock acid solution and curve B after 10 min.

One point which should be made clear about these pK_a determinations is the time factor. The spectral measurements being discussed here were made at a time not less than 30 s and not more than 5 min after adjustment of the pH of the stock solution. As will become apparent when kinetics are discussed, the equilibrium of eq 1 is established rapidly, and is complete before the lower limit mentioned above. The upper limit is necessary because there does follow a much slower transformation, also to be discussed later.

Presence of the "Intermediate" Stage. Figure 3 illustrates a further important spectral observation. Spectrum B of that figure is identical with the spectrum labeled "neutral-initial" of Figure 1, the spectrum which we have assigned to the "neutral" species in the acid–base equilibrium of eq 1. At the pH used in obtaining Figure 3, however, pH 8.3, this spectrum is obtained only after 5–10 min, the spectrum A obtained after 30 s being slightly different. The difference between the two cannot be due to incomplete reaction of the flavylium ion since in spectrum A there is no absorbance at the λ_{max} of the ion. We will use the term "intermediate" to describe these spectral changes. They follow

---

(7) Sondheimer, E. *J. Am. Chem. Soc.* **1953**, *75*, 1507.

(8) Jurd, L. *J. Org. Chem.* **1963**, *28*, 987.

(9) Jurd, L. *J. Food Sci.* **1964**, *29*, 16.

(10) Timberlake, C. F.; Bridle, P. *Nature (London)* **1966**, *212*, 518.

(11) Harper, K. A.; Chandler, B. V. *Aust. J. Chem.* **1967**, *20*, 731.

(12) Nilson, E. *Acta Chem. Scand.* **1967**, *21*, 1942.

(13) Brouillard, R.; Dubois, J. E. *J. Am. Chem. Soc.* **1977**, *99*, 1359.

(14) Brouillard, R.; Delaporte, B. *J. Am. Chem. Soc.* **1977**, *99*, 8461.

(15) Brouillard, R.; Delaporte, B.; Dubois, J. E. *J. Am. Chem. Soc.* **1978**, *100*, 6202.

(16) Jurd, L. *Tetrahedron* **1969**, *23*, 2367.

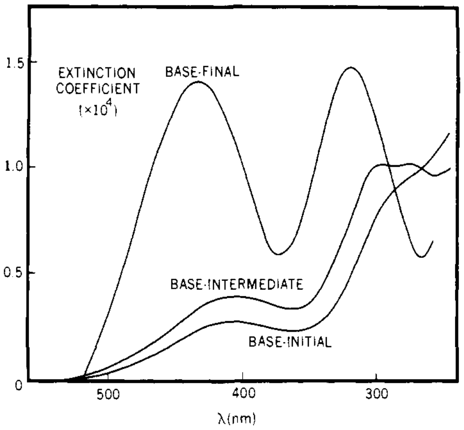


Figure 4. Spectra of 4′-methoxyflavylium perchlorate in 0.05 M NaOH. Curves were obtained after 20–40 s (initial), 30 min (intermediate), and 3 days (final). The spectrum labeled final is also observed with *trans*-2-hydroxy-4′-methoxychalcone in 0.05 M NaOH.

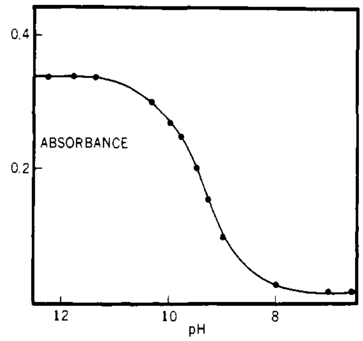


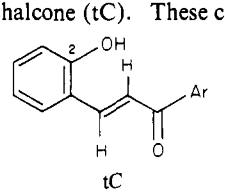
Figure 5. Absorbance at 410 nm of 6.4 × 10^{-5} M 4′-methoxyflavylium perchlorate, solutions measured 25 min after mixing of the stock acid solution. The curve drawn is the titration curve for an acid with pK_a = 9.43 and limiting absorbances of 0.34 in base and 0.01 in acid.

the rapid disappearance of F^+, but are themselves followed by a much slower reaction, the nature of which will be discussed later. We can also note that reacidification at any time during this "intermediate" stage rapidly and quantitatively regenerates flavylium ion.

Spectra and Acidity Constant in Base. Solutions of the three flavylium ions in more basic media are colored, even initially (Figure 4). The chief characteristics of the pH 8 behavior are, however, retained. There are three separate stages of spectral changes, the initial disappearance of F^+ generating the "initial" spectrum of Figure 4, and an "intermediate" change, with the system up to this point being completely reversible by reacidification. These are followed by a slow final change.

The change in the nature of the absorption spectra in proceeding from neutral pH to basic pH follows the behavior of a simple acid–base equilibrium (Figure 5). As will become apparent in the discussion, equilibrium is only reached after completion of the "intermediate" stage, and, since it is only with the 4′-methoxy system that this stage is well separate from the final stage, for a quantitative calculation we have limited ourselves to that system. We define the acidity constant as pK^a_{obsd}(base), and in the 4′-methoxy system this has a value of 9.43.

Conversion to *trans*-Chalcone. As has been alluded to on several occasions, there is a slow spectral change in solutions throughout the pH region, except in strong acids. The product of this reaction is a *trans*-2-hydroxychalcone (tC). These compounds have been prepared by separate synthesis, and their geometry has been verified by the characteristic trans coupling constants, J = 18 Hz, of the vinylic protons. Proof of this structure for the ultimate product in the hydration was obtained in one case, 4′-methoxyflavylium ion at pH 7, by isolating the product after 2 weeks standing, and finding this to be identical with the synthetic sample. In other cases we simply noted the identical nature of the absorption spectra of the chalcone and the final product of the flavylium ion hydrolysis (Figures 1 and 3). These absorption spectra, when obtained in neutral and acid solutions (pH <8.5), have peaks in the 300–400-nm region (see Figure 1 for an example), a behavior which appears to be characteristic of *trans*-2-hydroxychalcones.[16–18] In base the spectra of these compounds have a peak in the visible region (see Figure 3) associated with the ionization of the phenolic OH.



tC

This final reaction is irreversible, no flavylium ion being formed on reacidification to pH 1 of any solution allowed to stand for sufficient time to convert completely to *trans*-chalcone. This is obviously true also of a solution of the synthetic *trans*-chalcone in 0.1 M HCl. This applies not only to freshly prepared acid solutions, but also to solutions of tC in 0.1 M HCl left for 1 week. Even then no flavylium ion has appeared, the absorption spectrum of such a solution being the same as the spectrum of tC in a neutral solution. Flavylium ion does form in a rather slow reaction in solutions of *trans*-chalcone in more concentrated acids (30–70% $HClO_4$), and we are currently studying this in detail and will report on it at a later date.

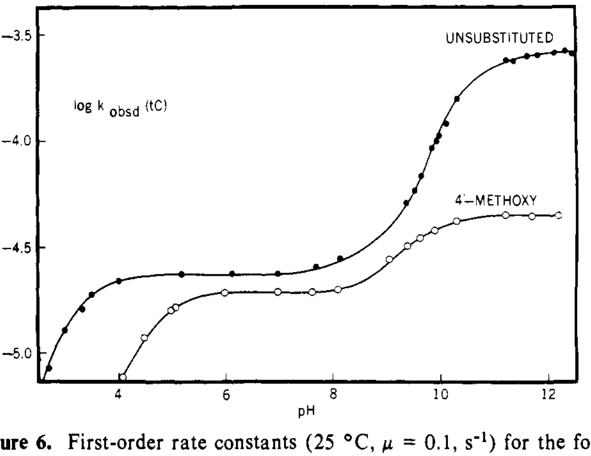


Figure 6. First-order rate constants (25 °C, μ = 0.1, s^{-1}) for the formation of *trans*-2-hydroxychalcones.

A kinetic study of the *trans*-chalcone forming reaction shows that it is first order at constant pH, with no catalysis by the components of the buffer. Figure 6 depicts the rate behavior as a function of pH for the 4′-methoxy and unsubstituted systems (4′-methyl was not studied in detail). The kinetic behavior in solutions of pH >6 is described by the equation

$$k^{\mathrm{pH}>6}_{\mathrm{obsd}}(\mathrm{tC}) = \frac{k^{\mathrm{base}}_{\mathrm{obsd}}(\mathrm{tC})K^{\mathrm{a}}_{\mathrm{obsd}}(\mathrm{base}) + k^{\mathrm{neut}}_{\mathrm{obsd}}(\mathrm{tC})[\mathrm{H}^+]}{K^{\mathrm{a}}_{\mathrm{obsd}}(\mathrm{base}) + [\mathrm{H}^+]} \tag{3}$$

$k^{\mathrm{base}}_{\mathrm{obsd}}(\mathrm{tC})$ and $k^{\mathrm{neut}}_{\mathrm{obsd}}(\mathrm{tC})$ are the rate constants in base and neutral solution, and pK^a_{obsd}(base) is a kinetic acidity constant. The value of the last constant for the 4′-methoxy system is the same as the spectroscopic acidity constant also measured for this system in base, so that we have used the same symbol. Our analysis will demonstrate that they have the same significance. A further decrease in rate is observed in more acidic solutions, this decrease occurring when the F^+–neutral equilibrium starts shifting toward F^+. Equation 4 accurately describes this behavior.

$$k^{\mathrm{pH}<6}_{\mathrm{obsd}}(\mathrm{tC}) = k^{\mathrm{neut}}_{\mathrm{obsd}}(\mathrm{tC})\left\{\frac{K^{\mathrm{a}}_{\mathrm{obsd}}(\mathrm{F}^+)}{K^{\mathrm{a}}_{\mathrm{obsd}}(\mathrm{F}^+) + [\mathrm{H}^+]}\right\} \tag{4}$$

---

(17) Jurd, L.; Horowitz, R. M. *J. Org. Chem.* **1961**, *26*, 2561.

(18) Jurd had originally assigned a cis structure to chalcone product of the hydration of some flavylium ions,[8] but subsequently revised this[16] on the basis of NMR evidence. It is also likely that the chalcones studied in ref 17 have a trans geometry.

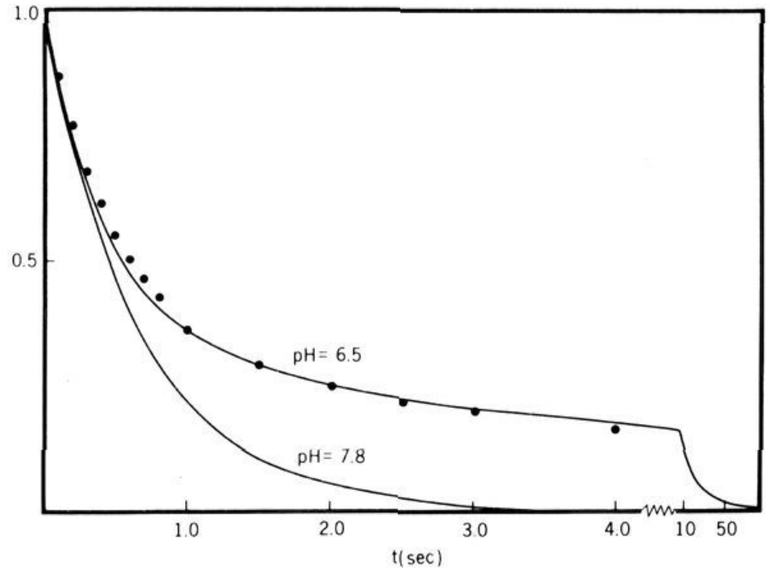


Figure 7. Disappearance of the 4′-methoxyflavylium ion as a function of time. The curves are experimental and plot A/A_0, where A and A_0 refer to the absorbance at 439 nm at times t and zero, respectively. The points at pH 6.5 are calculated values of $[F^+]_t/[F^+]_0$ (see Discussion).

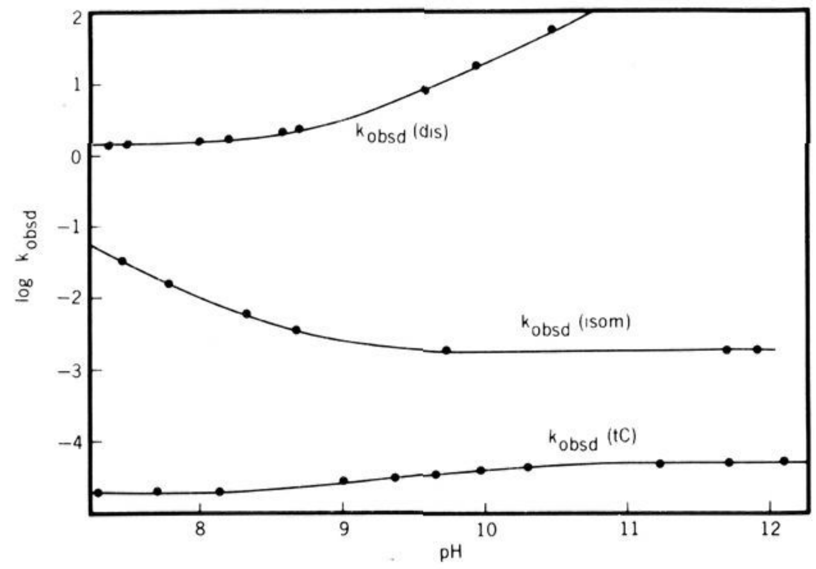


Figure 8. First-order rate constants (25 °C, $\mu = 0.1$, s^{-1}) associated with spectral changes occurring in solutions of 4′-methoxyflavylium perchlorate. For the top two curves, k_{obsd} values are those obtained on extrapolation to zero buffer concentration.

$k^{neut}_{obsd}(tC)$ has the same meaning as above, and $K^a_{obsd}(F^+)$ is the acidity constant of F^+ determined previously.

Kinetics of Disappearance of Flavylium Ion in Neutral and Base Solution. Although we have referred to the disappearance of flavylium ion in neutral and base solutions as being rapid, it can be easily seen in a stopped-flow spectrophotometer. Figure 7 depicts the absorbance as a function of time of the 4′-methoxyflavylium ion in a solution of pH 6.5. This pH is 2.2 units greater than the $pK^a_{obsd}(F^+)$, and F^+ therefore is converted to the colorless neutral species to an extent greater than 99% in the solution. Its disappearance, however, does not obey first-order kinetics. There appears to be two not quite separate kinetic phases, a rapid phase in which the absorbance decreases to some intermediate value, followed by a slower phase in which it further decreases to almost zero.

This biphasic behavior is not seen in all solutions. As the pH is increased, the relative absorbance change in the second phase diminishes, and eventually a single first-order curve is observed, such as that at pH 7.8 in Figure 7. Similar behavior is observed for the other two ions, the generalization being that first-order kinetics are only found when the solution has a pH at least 3.5 log units greater than the $pK^a_{obsd}(F^+)$.

Where first-order kinetics are observed, the rate law of eq 5 is found, as shown in Figure 8 for the 4′-methoxy compound.

$$k_{obsd}(dis) = k^{w}_{obsd}(dis) + k^{OH}_{obsd}(dis)[OH^-] + k^{B}_{obsd}(dis)[B] \quad (5)$$

The last term is due to the base component of the buffer, and is relatively unimportant, in our buffers accounting for 20% of $k_{obsd}(dis)$ at the most. The first two terms show the presence of a pH-independent reaction and a reaction first order in hydroxide ion.

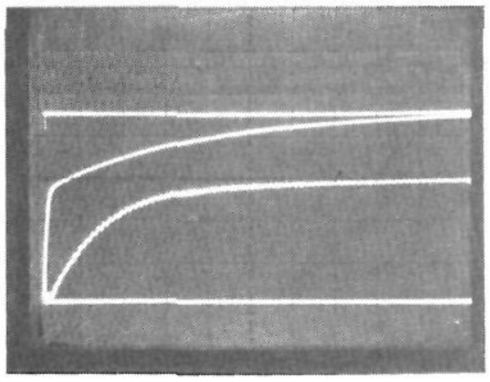


Figure 9. Stopped-flow oscilloscope traces obtained when a solution of 4′-methoxyflavylium perchlorate in dilute Tris buffer ([buffer] = 0.005, pH 7.25) is mixed with dilute HCl to give a solution with final pH 2.50. The wavelength is 440 nm, with one vertical division = 0.2 optical density (OD) units. The bottom line corresponds to zero OD, the top line to the stable final OD. The bottom curve was recorded at 20 ms/div, the top curve at 500 ms/div.

Kinetics of Intermediate Spectral Change. The intermediate change is first order in a given solution, and obeys the rate law

$$k_{obsd}(isom) = k^{H}_{obsd}(isom)[H^+] + k^{w}_{obsd}(isom) + k^{HA}_{obsd}(isom)[HA] \quad (6)$$

(see again Figure 8 for results in the 4′-methoxy system). The last term is due to the acid component of the buffer; its contribution is relatively unimportant. The first two terms show a hydronium ion dependent reaction and a pH-independent reaction.[19]

It can be seen in Figure 8 that on proceeding to acid pH the rate of the intermediate stage becomes rapid, because of the H^+-dependent reaction. This accounts for this stage not being seen in constructing the "neutral-initial" spectral curve of Figure 1; it is complete before the spectrum was taken. This will also be true throughout the acidic pH region. Thus, in the determination of $pK^a_{obsd}(F^+)$, the "neutral" species refers to whatever is present after the intermediate stage is complete.

Kinetics of F^+ Formation on Reacidification of Neutral Solutions. After Completion of Intermediate Stage. Like the disappearance of flavylium ion in neutral and base solutions, its reappearance on reacidification of these solutions, although rapid, can be seen in a stopped-flow spectrophotometer. We will start with the situation which arises with solutions near neutrality which have been left for a time sufficient that the intermediate stage is complete. As can be seen in Figure 9, the surprising observation is made that the reappearance of F^+ from these solutions occurs in two separate phases, a very rapid phase in which the absorbance of F^+ starts near zero and rises to some intermediate value, followed by a slower phase in which the absorbance further increases to its stable final value corresponding to the quantitative regeneration of the ion. Both phases obey first-order kinetics in a given acid, and, except in very dilute acids, the two are sufficiently different in rate that accurate first-order rate constants can be obtained for each. These rate constants are shown as a function of acidity in Figures 10 and 11, and obey the rate laws

$$k_{obsd}(F^+\text{-slow}) = k^{H}_{obsd}(F^+\text{-slow})[H^+] + k^{w}_{obsd}(F^+\text{-slow}) \quad (7)$$

$$k_{obsd}(F^+\text{-fast}) = k^{H}_{obsd}(F^+\text{-fast})[H^+] \quad (8)$$

The slow phase reveals reactions which are H^+ dependent and pH independent, while the fast phase appears only to involve an H^+-dependent reaction, its rate constants extrapolating, within experimental error, to zero at zero H^+ concentration. Our later analysis will show that there is associated with this phase a pH-independent reaction, but its rate constant is very small, so that the reaction cannot be seen at low pH. For the 4′-methoxy compound rate constants were obtained for the slow phase in very dilute acids and appear to start increasing again. Our later analysis will also show that this is real.

---

[19] A value of the pH-independent rate constant $k^{w}_{obsd}(isom)$ was only obtained for the 4′-methoxy system, since with the other two ions values of $k_{obsd}(isom)$ in base solutions, where the pH-independent process is important, cannot be accurately measured because of interference from the *trans*-chalcone forming process.

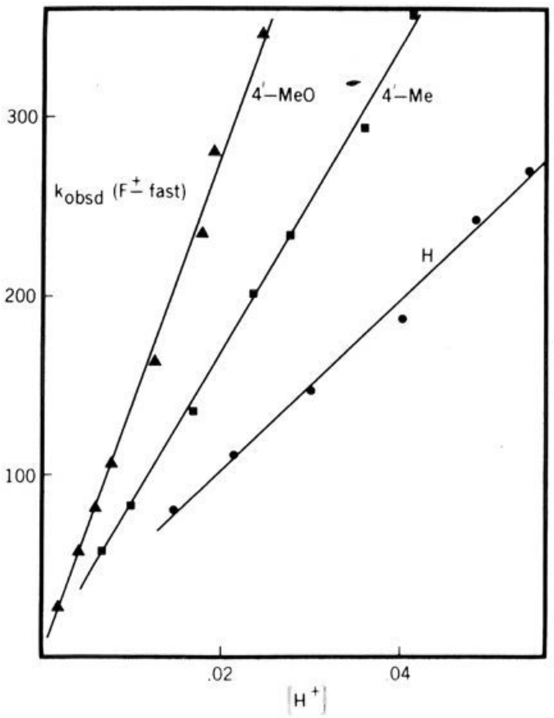


Figure 10. First-order rate constants (25 °C, $\mu = 0.1$, s^{-1}) for the appearance of flavylium ion in acid solutions, fast kinetic phase.

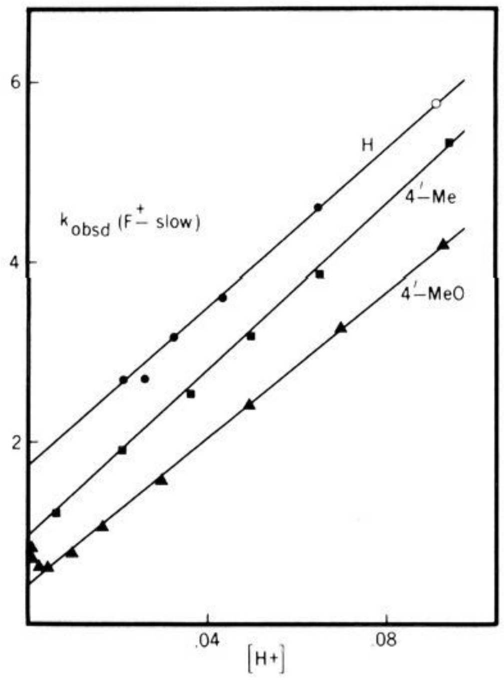


Figure 11. First-order rate constants (25 °C, $\mu = 0.1$, s^{-1}) for the appearance of flavylium ion in acid solutions, slow kinetic phase.

We also define a quantity ΔOD(sloow)/ΔOD(fast) equal to the relative absorbance change observed in the two phases. This ratio is dependent on the substituent in the flavylium ion, but otherwise does not change (a) with changing pH of the reacidified solution, (b) with changing pH of the neutral solution up to about 8.5, and (c) with changing time of standing of the neutral solution. In the last case, the actual amount of flavylium ion produced on reacidification decreases with time as *trans*-chalcone is formed irreversibly in the neutral solution, but the relative absorbance change is always the same.

Flavylium Ion Formation on Reacidification of Neutral Solutions. During the Intermediate Stage. In these experiments we concentrated on preparing the neutralized solution, loading it into the stopped-flow apparatus, and reacidifying as quickly as possible, our intent being to catch the system before the intermediate stage is complete. A typical result is shown in Figure 12. The two first-order reactions observed in Figure 9 can still be seen, and have the same rate constants and same relative OD changes. However, when the reacidification is done quickly, the initial absorbance is not zero; there is already flavylium ion present. This initial absorbance decreases with time, at a rate about equal to that associated with the intermediate stage (k_{obsd}(isom)), and within 10–20 min, when the intermediate stage is complete, the initial absorbance has dropped to zero, and the oscilloscope traces become identical with those of Figure 9. Although we will delay our explanation until the Discussion, we can point out here that the initial absorbance is not due to incomplete hydrolysis of the flavylium ion in the neutral solution, since the half-life of the ion is less than 1 s in these solutions (0.3 s for the system of Figure 12), and yet the initial absorbance is seen even after minutes.

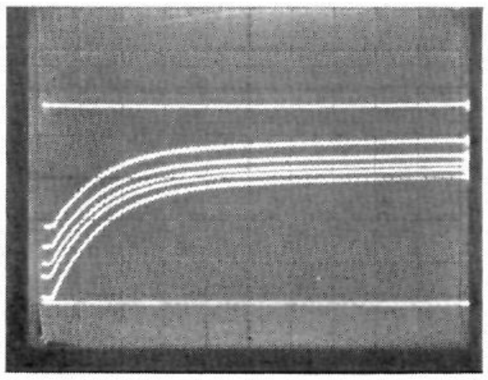


Figure 12. Stopped-flow oscilloscope traces obtained when a solution of 4′-methoxyflavylium perchlorate in dilute Tris buffer ([buffer] = 0.005, pH 8.3) is mixed with dilute HCl to give a solution with a final pH 2.45. The wavelength is 440 nm, with 0.2 OD unit/div and 20 ms/div. The bottom line corresponds to zero OD; the top line is the stable final OD. Curves were obtained on repeated mixings in the stopped-flow apparatus, 40 s, 70 s, 100 s, 140 s, and 20 min, starting at the top, after the neutralized solution was prepared.

Flavylium Ion Formation on Reacidification of Base Solutions. Reacidification of base solutions of flavylium ions in which the intermediate stage is complete results in oscilloscope traces similar to those of Figure 9 in that the initial absorbance starts near zero, and there are two kinetic phases, the rate constants for which are the same as those obtained starting with neutral solutions. A difference, however, is that with base solutions most of the absorbance increase occurs in the slow phase.

Reacidification of base solutions before the intermediate stage is complete results in oscilloscope traces like those in Figure 12, in which there is a significant initial absorbance which decreases in magnitude with the time of standing of the solution. Of the remaining absorbance increase most is accounted for in the slow phase.

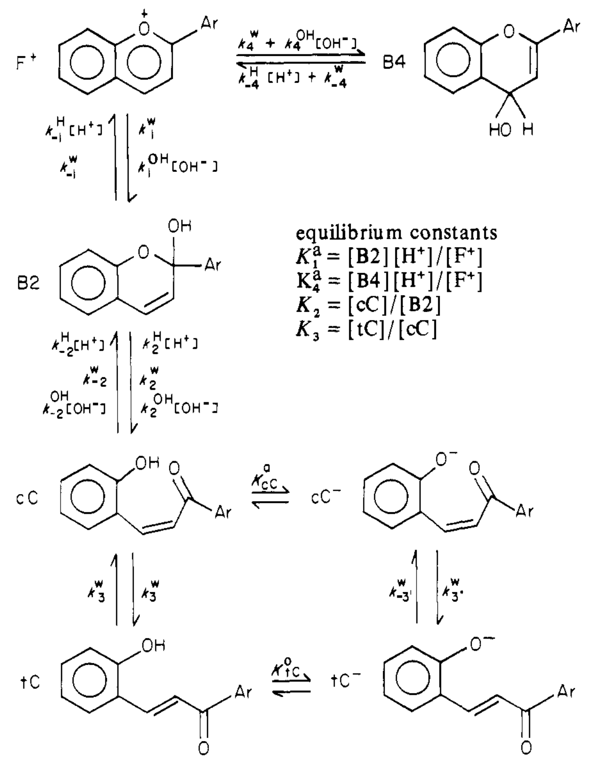
Reacidification of a Solution Containing Only *trans*-2-Hydroxychalcone. A 1.4×10^{-4} M solution of 4′-methoxyflavylium perchlorate in 0.002 M NaOH was left for 7 days, a time corresponding to 20 half-lives of the reaction which forms *trans*-chalcone, and then reacidified to pH 2.5 in the stopped-flow spectrophotometer. The resulting solution does have a small absorbance (0.05 OD unit) at the λ_{max} of the flavylium ion, but this is not unexpected since the spectrum of the *trans*-chalcone tails into the visible region, and this absorbance can be thus attributed to the *trans*-chalcone. What we were looking for was a first-order increase in absorbance at this wavelength corresponding to formation of flavylium ion. There is none.

The intention of this experiment was to see if any of the compound which forms F$^+$ on acidification is still present in equilibrium with the *trans*-chalcone. As a control to evaluate the sensitivity of the approach, to a portion of the fully transformed solution a fresh sample of 4′-methoxyflavylium perchlorate, concentration 1.6×10^{-7} M, was added. Immediate reacidification of this solution does result in an observable first-order increase in absorbance at 440 nm of 0.012 OD unit, with the same rate constant as found on reacidification of more concentrated freshly prepared solutions. This observation is important since it establishes that the observation is not an artifact. The absorbance change corresponds to the production of 1.4×10^{-7} M F$^+$ ($\epsilon = 4.2 \times 10^4$, path length = 2 cm), accounting reasonably well for that which was originally added. We feel that an absorbance change of 0.002 could be observed in such an experiment. For the fully transformed solution, therefore, we place an upper limit on the concentration of the species which forms F$^+$ on acidification of 2.4×10^{-8} M. Thus its equilibrium concentration is less than 1/5000 that of *trans*-2-hydroxychalcone (actually *trans*-chalcone anion in 0.002 M NaOH).

Discussion

General Scheme. In the only previous report involving any of the ions of this investigation, Jurd,[16] in a study admittedly not

Scheme I



equilibrium constants
$K_1^a = [B2][H^+]/[F^+]$
$K_4^a = [B4][H^+]/[F^+]$
$K_2 = [cC]/[B2]$
$K_3 = [tC]/[cC]$

detailed, suggested for the 4′-methoxyflavylium ion a very simple hydrolysis. He observed conversion in neutral solutions to a stable, colorless species, and tentatively assigned to that species a pseudobase structure. We also observe these colorless, neutral solutions, although we find that they are not indefinitely stable, being converted eventually to solutions of *trans*-2-hydroxychalcone. Our results moreover suggest that the chemistry is far more complex than that suggested by Jurd.

In our discussion we will be referring to the species and reaction stages defined in Scheme I. We will be presenting arguments that all of the ions and molecules of this scheme are present at some time under some conditions, and moreover our kinetic analysis will furnish all of the rate constants and equilibrium constants of this scheme, with the exception of K_3, k_{-3}^w, and $k_{-3'}^w$, for which we only have lower or upper limits at this time. It should be noted that the rate and equilibrium constants of Scheme I are defined with a number subscript to distinguish these from the observed parameters. Numerical values of the parameters of Scheme I are to be found in Table II.

Our proposals will be supported mainly by a series of kinetic arguments. These arguments are complex in many instances, and taken individually it is probably true that alternative interpretations could be given. Taken collectively, however, our interpretation produces a self-consistency, which, in our opinion, cannot be obtained with other explanations.

Equilibrium between B2 and cC and Conversion to F⁺. We will consider first the question as to what is present in a hydrolyzed solution of F⁺ in the neutral pH region, after the stage which we have labeled intermediate is complete. Our proposal is that this solution contains a rapidly equilibrating mixture of B2 and cC. Our first arguments to support this proposal are based on the biphasic kinetics observed in the stopped-flow experiments when such solutions are reacidified (Figure 9) and re-form F⁺. We suggest that the first kinetic phase represents the formation of F⁺ from the B2 of the mixture, while the second phase represents the formation of F⁺ from cC. The latter conversion must proceed via B2, but since in these acids B2 is converted to F⁺ more rapidly than it forms from cC the rate-determining step[20] is the conversion to B2. In other words, the rapid acidification of this neutral solution traps the pseudobase, converting it to F⁺ before virtually any cC has a chance to react. The solution at this point contains F⁺ and cC, and the latter then undergoes its slower reaction resulting in the formation of the remainder of the ion.

Table II. Rate Constants and Equilibrium Constants for the Transformations of 4′-Substituted Flavylium Ions (25 °C, μ = 0.1)[a]

constant	4′-MeO	4′-Me	H
pK_1^a	4.47	3.65	3.01
k_1^w, s^{-1}	0.47	1.88	4.59
k_1^{OH}, M^{-1} s^{-1}	1.2×10^5	3.5×10^5	4.8×10^5
k_{-1}^H, M^{-1} s^{-1}	1.38×10^4	8.4×10^3	4.7×10^3
k_{-1}^w, s^{-1}	3.5×10^{-5}	1.6×10^{-5}	4.9×10^{-6}
pK_4^a	6.46	5.54	4.93
k_4^w, s^{-1}	0.91	4.02	8.1
k_4^{OH}, M^{-1} s^{-1}	6.0×10^4	1.7×10^5	2.4×10^5
k_{-4}^H, M^{-1} s^{-1}	2.6×10^6	1.4×10^6	6.9×10^5
k_{-4}^w, s^{-1}	1.7×10^{-3}	5.9×10^{-4}	2.0×10^{-4}
K_2	0.50	0.16	0.06
k_2^H, M^{-1} s^{-1}	19.5	7.0	2.5
k_2^w, s^{-1}	0.23	0.15	0.11
k_2^{OH}, M^{-1} s^{-1}	0.7×10^{10}	0.8×10^{10}	0.6×10^{10}
k_{-2}^H, s^{-1}	39	44	42
k_{-2}^w, s^{-1}	0.46	0.96	1.78
k_{-2}^{OH}, M^{-1} s^{-1}	1.3×10^{10}	5.2×10^{10}	9.4×10^{10}
k_3^w, s^{-1}	5.8×10^{-5}		4.1×10^{-4}
$k_{3'}^w$, s^{-1}	4.6×10^{-5}		2.6×10^{-4}
pK_{cC}^a	9.00 (kinet)		9.03 (kinet)

[a] See Scheme I for definitions.

Our initial arguments in support of this proposal are based on the effects of the 4′ substituent. If the preceding analysis is correct, the changes in absorbance in the fast phase and slow phase are proportional to the concentrations of B2 and cC in the neutral solution immediately before acidification. Assuming that the two are in equilibrium we can write

$$\Delta OD(slow)/\Delta OD(fast) = [cC]/[B2] = K_2 \qquad (9)$$

That B2 and cC are in equilibrium will be demonstrated shortly, although this fact is fairly evident from the constancy of the ΔOD ratio for a given ion as conditions are varied. As is generally observed for a carbonyl addition reaction of this type,[21] the equilibrium between B2 and cC is expected to shift more toward cC with increased electron donation in Ar, due to the interaction between the carbonyl group and the substituent. Precisely such a trend is seen in the values of K_2, which is largest in the 4′-methoxy system and smallest in the unsubstituted system.

The H⁺-catalyzed fast kinetic phase is proposed to represent the H⁺-catalyzed conversion of B2 to F⁺, so that we can write

$$k_{obsd}^H(F^+\text{-fast}) = k_{-1}^H \qquad (10)$$

The acceleration by electron donors (negative ρ[22]) observed for this phase is consistent with this interpretation, the ion and the transition state leading to it being stabilized by such substituents. The kinetics of the slow phase are proposed to represent cyclization of cC to B2, so that we can write

$$k_{obsd}^H(F^+\text{-slow}) = k_{-2}^H \qquad (11)$$

$$k_{obsd}^w(F^+\text{-slow}) = k_{-2}^w \qquad (12)$$

In contrast to the fast phase neither of these rate constants shows an acceleration by electron donors. This fact suggests immediately that the slow phase refers to some process in which F⁺ is formed

---

[20] It can be demonstrated by using using the rate constants derived in our analysis that B2, once formed, does not revert back to cC ($k_{-1}[H^+] > k_2^H[H^+] + k_2^w$).

[21] Hine, J. "Structural Effects on Equilibria in Organic Chemistry"; Wiley: New York, 1975; pp 257–265.

[22] We hesitate to actually list numerical values of ρ, since only three substituents are involved, and two of these can interact conjugatively with the reaction center.

after the rate-determining step. Moreover, the trends observed, a $\rho \approx 0$ for the H^+-catalyzed reaction and a positive ρ for the pH-independent reaction, are those that might be expected of a carbonyl addition reaction of the cC → B2 type. The H^+-catalyzed addition of a nucleophile to a carbonyl group is generally associated with a ρ value near zero, as, for example, in the acid-catalyzed hydrolysis of esters.[23] This is due to a cancellation of effects, electron donation favoring H^+ transfer but retarding the addition reaction. In a noncatalyzed addition, however, only the latter effect is important and the ρ value should be positive.[24]

Before proceeding to further evidence, we can note that several other constants of Scheme I are obtainable from the constants derived from eq 9–12. Knowledge of the equilibrium constant K_2 and the rate constants k_{-2}^{H} and k_{-2}^{w} provides[25] automatically the rate constants k_2^{H} and k_2^{w}. This incidentally also provides the minimum rate of equilibration of B2 and cC, as the sum of the forward and reverse pH-independent constants, $k_{-2}^{w} + k_2^{w}$. Even these indicate half-lives of equilibration of the order of seconds.

The above interpretation also asserts that the neutral species in equilibrium with F^+ in more acidic solutions is not one chemical substance but in fact an equilibrating mixture of B2 and cC, so that we are dealing with the equilibrium

$$F^+ \underset{}{\overset{K^a_1}{\rightleftharpoons}} B2 \overset{K_2}{\rightleftharpoons} cC \tag{13}$$

The observed acidity constant is then given by

$$K^a_{obsd}(F^+) = \frac{([B2] + [cC])[H^+]}{[F^+]} = K^a_1(1 + K_2) \tag{14}$$

Since K_2 has been evaluated, we can calculate K^a_1, the true acidity constant for pseudobase formation. Furthermore, using the values of K^a_1 and k^H_{-1} (eq 10), we can calculate k^w_1, the rate constant for reaction of F^+ and water.[26]

Evidence for *cis*-Chalcone. Ionization in Base. Additional evidence for the presence of *cis*-chalcone is afforded by observations made when the hydrolysis of the flavylium ion is carried out in more basic solutions. In particular the kinetic reacidification experiments of such solutions reveal that the slow kinetic phase almost completely dominates, in marked contrast to the behavior with neutral solutions. This can be explained by a shifting in base of the equilibrium between the pseudobase form and the *cis*-chalcone form, owing to the ionization of the latter.

$$B2^- \overset{1/K^a_{B2}}{\rightleftharpoons} B2 \overset{K_2}{\rightleftharpoons} cC \overset{K^a_{cC}}{\rightleftharpoons} cC^- \tag{15}$$

Evidence for this assertion comes from the fact that there is an acid–base equilibrium observed spectroscopically in base solutions of F^+, with the new peak which is observed in the visible region in base being assigned to the ionized *cis*-chalcone. The observed acidity constant can be converted to the true ionization constant of the *cis*-chalcone by recognizing that the neutral component of the equilibrium is an equilibrating mixture of B2 and cC.

$$K^a_{obsd}(\text{base}) = \frac{[cC^-][H^+]}{[B2] + [cC]} = \frac{K_2}{1 + K_2}K^a_{cC} \tag{16}$$

The value of pK^a_{cC} calculated for *cis*-2-hydroxy-4′-methoxychalcone is 8.93. This is of the order expected of a phenol ionization.

One assumption has been made in the derivation of eq 16, namely, that the hydroxyl group of B2 ionizes with greater difficulty. This assumption is probably valid. The pK_a values of a number of hemiacetals or carbonyl hydrates have been measured or estimated, and they generally lie between 12 and 13.[27–31] This will mean that B2 would exist in ionized form in strongly basic solutions. However, the much greater acidity of the phenolic OH group of cC will shift the equilibrium well toward the *cis*-chalcone before this happens. If it is assumed that $pK^a_{B2} = 12$, the values of pK^a_{cC} and K_2 can be used to calculate the equilibrium constant for the system $B2^- \rightleftharpoons cC^-$. The value so obtained is 5.6×10^2 (for 4′-methoxy).

Rate of Equilibration of B2 and cC in Neutral Solutions. The displacement of the pseudobase–*cis*-chalcone equilibrium in base provides a way of obtaining their rate of equilibration in neutral solutions, by mixing the base solution in a stopped-flow spectrophotometer with appropriate neutral buffers. The principle of this approach, which we have also successfully applied to study the breakdown of benzaldehyde hydrates[24b] and cyclic hemiacetals,[31] is that B2 and cC are not at equilibrium in the neutral solution immediately after mixing because of the displacement in base, and the return to equilibrium is then monitored spectroscopically. Indeed, when pH 11.5 solutions of the three flavylium ions are mixed with acetate buffers of pH 4.5–6.5, a small but significant first-order decrease in absorbance at 290–300 nm[32] is observed. First-order rate constants obey the rate law

$$k_{obsd}(B2 \rightleftharpoons cC) = k^{OH}_{obsd}(B2 \rightleftharpoons cC)[OH^-] + k^{OAc}_{obsd}(B2 \rightleftharpoons cC)[OAc^-] \tag{17}$$

showing catalysis of the equilibration only by hydroxide ion and acetate ion (general base catalysis) in this pH region.[33] It can be recalled that values of $k_{obsd}(F^+\text{-slow})$, which we propose to also represent the conversion of cC to B2, show some sign of an upswing with increasing pH even at pH 3–3.5 (Figure 11). This can now be explained as the onset of the hydroxide ion catalysis of this reaction.[34]

Conversion of the empirical rate constant of eq 17 to the constants of Scheme I is carried out by recognizing that the former refers to an approach to equilibrium and therefore is given by the sum of forward and reverse rate constants:

$$k^{OH}_{obsd}(B2 \rightleftharpoons cC) = k_2^{OH} + k_{-2}^{OH} \tag{18}$$

Since the ratio k_2^{OH}/k_{-2}^{OH} is equal to K_2, which has been evaluated, the individual constants can be calculated.

The enormous values of these rate constants, both k_2^{OH} and k_{-2}^{OH} being in the range 10^{10}–10^{11} M^{-1} s^{-1}, may seem surprising. We can, however, refer to two somewhat similar systems where the hydroxide ion catalyzed breakdown of a carbonyl adduct has a rate in the same range (eq 19). Values of k^{OH} for the reaction of eq 19a have been measured[35,36] as 5–6 × 10^{10} M^{-1} s^{-1}, and a kinetic analysis shows[37] that values of k^{OH} for eq 19b must be greater than 5 × 10^{10} M^{-1} s^{-1}. We can also quote one example[31]

---

[23] Kirby, A. J. In "Comprehensive Chemical Kinetics", Bamford, C. H., Tipper, C. F. H., Eds.; Elsevier: Amsterdam, 1972; Vol. 10.

[24] (a) As, for example, in the noncatalyzed hydration of substituted benzaldehydes.[24b] (b) McClelland, R. A.; Coe, M., to be published.

[25] $K_2 = k_2^{H}/k_{-2}^{H} = k_2^{w}/k_{-2}^{w} = k_2^{OH}/k_{-2}^{OH}$.

[26] $K^a_n = k^w_n/k^H_{-n} = k^{OH}_n K_w/k^w_{-n}$, for both $n = 1$ and 4.

[27] (a) Isbell, H. S.; Pigman, W. *Adv. Carbohydr. Chem.* **1969**, *24*, 13. (b) Pigman, W.; Isbell, W. S. *Ibid.* **1968**, *23*, 11.

[28] Los, J. M.; Simpson, L. B. *Recl. Trav. Chim. Pays-Bas* **1956**, *75*, 267.

[29] Greenzaid, P. *J. Org. Chem.* **1973**, *38*, 3164.

[30] Sayer, J. M. *J. Org. Chem.* **1975**, *40*, 2545.

[31] Harron, J.; McClelland, R. A.; Thankachan, C.; Tidwell, T. T., *J. Org. Chem.*, submitted.

[32] We feel that neutral *cis*-2-hydroxychalcones have absorption spectra with a λ_{max} near 300 nm (see Figures 1 and 3). A decrease in absorbance at this wavelength is observed in the $B2 \rightleftharpoons cC$ equilibration experiment as the excess cC is converted to B2. In contrast, an increase in absorbance at this wavelength is observed in the intermediate stage, a stage which we propose to involve formation of cC (in equilibrium with B2). The lower λ_{max} of the *cis*-2-hydroxychalcone as compared to the trans isomer indicates to us that the cis compound is not planar.

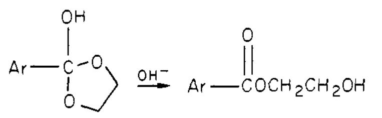
[33] This is due to the fact that the pH-independent rate constant which would be equal to $k_2^{w} + k_{-2}^{w}$ is simply too small to be significant. Unfortunately these experiments cannot be pushed to more acidic solutions because appreciable quantities of F^+ start being formed, and this reaction interferes with the kinetics of isomerization.

[34] (a) It can be noted that for the interconversion of cC and F^+ there is a change in rate-determining step between high pH and low pH, with the interconversion of cC and B2 being rate limiting at low pH and the interconversion of F^+ and B2 rate limiting at high pH. A similar change in rate-determining step is observed in the related lactonization of coumarinic acid.[34b] (b) Herschfield, R.; Schmir, G. L. *J. Am. Chem. Soc.* **1973**, *95*, 7360.

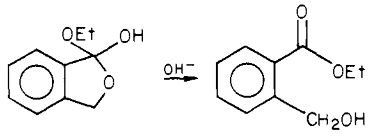
[35] Ahmad, M.; Bergstrom, R. G.; Cashen, M. J.; Chiang, Y.; Kresge, A. J.; McClelland, R. A.; Powell, M. F. *J. Am. Chem. Soc.* **1979**, *101*, 2669.

[36] McClelland, R. A.; Ahmad, M. *J. Org. Chem.* **1979**, *44*, 1855.

[37] McClelland, R. A.; Alibhai, M., manuscript in preparation.

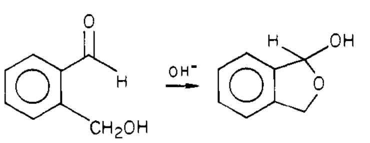


(19a)



(19b)

of a rapid ring closure, eq 20, which which $k^{OH} = 9 \times 10^7\ M^{-1}$



(20)

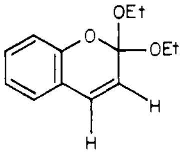
s^{-1}.

Mechanistically these reactions probably proceed via the anions of the species involved,[38] as in the equation

$$cC \rightleftharpoons cC^- \rightleftharpoons B2^- \rightleftharpoons B2 \qquad (21)$$

The large rate constants, which are near the diffusion limit,[39] suggest that one of the proton-transfer steps could be rate determining. Further experiments are currently in progress examining this question.

NMR Spectrum of B2. Although there are several additional kinetic arguments that support our proposal that the pseudobase B2 is the other species present in the equilibrium, we have been able to obtain its NMR spectrum,[40] and feel that this is sufficient evidence for our purposes. The experiment involved immediately extracting with ether a solution of flavylium ion at pH 7. Removal of the ether results in an unstable,[41] pale yellow solid or liquid which gives an NMR spectrum such as that depicted in Figure 13 for the parent system. This spectrum has two doublets, J = 9.5 Hz, at δ 5.80 and 6.60, integrating 1:1, with a complex set of peaks at δ 6.9–7.7 integrating to 9. The doublets can be assigned to the vinylic protons of the pseudobase. These have a coupling constant appropriate for the cis arrangement in that compound.[42] Further corroboration for this assignment comes from comparison with coumarin diethyl acetal,[43] which has a very similar spectrum, the vinylic protons appearing at δ 5.72 and 6.75, with J = 10 Hz.



Formation of B4 as Kinetic Product. The first-order kinetics observed for F^+ disappearance at high pH (eq 5) reveal the presence of a pH-independent reaction and a reaction first order in OH^-. The former must represent the reaction of F^+ with a solvent water molecule, and the latter the direct combination of F^+ and OH^-. Since two pseudobases in principle can form in each reaction

$$B4 \xleftarrow{k^w{}_4 + k^{OH}{}_4[OH^-]} F^+ \xrightarrow{k^w{}_1 + k^{OH}{}_1[OH^-]} B2 \qquad (22)$$

the observed first-order rate constant for F^+ disappearance is given by

$$k_{obsd}(\text{dis}) = (k_1^w + k_4^w) + (k_1^{OH} + k_4^{OH})[OH^-] \qquad (23)$$

Thus the empirical rate constants of eq 5 are given by

$$k_{obsd}^w(\text{dis}) = k_1^w + k_4^w \qquad (24)$$

$$k_{obsd}^{OH}(\text{dis}) = k_1^{OH} + k_4^{OH} \qquad (25)$$

Values of k_1^w in eq 24 have been obtained from independent results (see discussion near eq 14). If B2 were to be the only product of hydration, values of k_{obsd}^w(dis) should be the same as these, within experimental uncertainty. The values of k_{obsd}^w(dis) are, however, about three times greater. Thus a significant quantity of B4, about two-thirds of the product mixture, is formed in the initial hydration with H_2O. The remaining one-third consists of an equilibrium mixture of B2 and cC, this mixture being obtained rather than B2 itself since the rate of equilibration of these two is far more rapid than the rate of F^+ hydration.

This proposal of an initial formation of a second pseudobase form is consistent with several other observations. The intermediate spectral change, for example, can be interpreted as an isomerization of these initial products, the products of kinetic control, to an equilibrium situation, or to the products of thermodynamic control (using the latter in a short term sense only since tC is in fact the ultimate product of thermodynamic control). If the assumption is made that only the rapidly equilibrating mixture of B2 and cC is present after isomerization, the kinetic system

$$B4 \underset{k^w{}_4 + k^{OH}{}_4[OH^-]}{\overset{k^H{}_{-4}[H^+] + k^w{}_{-4}}{\rightleftharpoons}} F^+ \xrightarrow{k^w{}_1 + k^{OH}{}_1[OH^-]} B2 \overset{\text{fast}}{\rightleftharpoons} cC \qquad (26)$$

can be written, where F^+ is an intermediate in the isomerization, but is present only in small stationary state amounts at the acidities involved. The empirical rate constants for the isomerization (eq 6) are then given by[44]

$$k_{obsd}^H(\text{isom}) = k_{-4}^H\left(\frac{k_1^w}{k_1^w + k_4^w}\right) \qquad (27)$$

$$k_{obsd}^w(\text{isom}) = k_{-4}^w\left(\frac{k_1^{OH}}{k_1^{OH} + k_4^{OH}}\right) \qquad (28)$$

In eq 27, both k_1^w and k_4^w have been obtained previously, so that k_{-4}^H can be calculated. Having both k_{-4}^H and k_4^w, their ratio[26] provides pK_4^a, the acidity constant for reversible formation of the pseudobase **B4** from F^+. Self-consistency is introduced into the analysis since the equilibrium constant for isomerization of the two pseudobases, $K(B4 \rightleftharpoons B2)$, is also calculable, as K_1^a/K_4^a. Values of this ratio are about 100 for each flavylium ion, showing that little **B4** is present after equilibration. It is a kinetic product of hydration only.

The apparent initial concentration of F^+ seen in certain reacidification experiments (Figure 12, for example) can also be accounted for, since this occurrence is directly associated with an incomplete intermediate stage and therefore the presence of B4 in the hydrated solution. Our explanation is that this initial concentration is the F^+ which has formed from B4 on reacidification. It appears as an initial absorbance, and the actual formation of F^+ cannot be seen, since the rate in acid of the conversion of B4 to F^+ is such that its observation is beyond the capability of even the stopped-flow technique. For example, at pH 2.45, the pH of Figure 12, k_{obsd} for the reaction B4 → F^+, calculated as $k_{-4}^H[H^+]$, is $9.2 \times 10^3\ s^{-1}$. A reaction of this velocity is 99.99% complete before an observation is made in our stopped-flow apparatus, which has a dead time of 0.002 s.

What we propose, in other words, is that the appearance of F^+ in Figure 12 is triphasic, with a very rapid hidden phase as B4 converts to F^+, followed by the two phases commented on previously representing the successive conversion of B2 and cC to

---

[38] Funderburk, L. H.; Aldwin, L.; Jencks, W. P. *J. Am. Chem. Soc.* **1978**, *100*, 5444.

[39] Eigen, M. *Angew. Chem., Int. Ed. Engl.* **1964**, *3*, 1.

[40] Despite a number of attempts we have been unable to date to obtain an NMR spectrum of the *cis*-chalcone. One possible reason for this is that the B2 ⇌ cC equilibrium shifts toward the less polar B2 in the organic solvents used to obtain the spectra.

[41] Any attempt to purify this results in its conversion to a viscous, green oil. This also occurs on standing for several hours.

[42] Bunting, J. W. *Adv. Heterocycl. Chem.* **1979**, *25*, 1.

[43] McClelland, R. A.; Somani, R.; Kresge, A. J. *Can. J. Chem.* **1979**, *57*, 2260.

[44] Actually $k_{obsd}(\text{isom}) = (k_{-4}^H[H^+] + k_{-4}^w)(k_1^w + k_1^{OH}[OH^-])/(k_1^w + k_4^w + k_1^{OH}[OH^-] + k_4^{OH}[OH^-])$ and eq 27 and 28 are obtained as limits.

F^+. The relative optical density changes in each phase are proportional to the concentrations of the three species immediately before acidification. The amount of the phase due to B4 decreases with the time of standing of the neutral solution, eventually completely vanishing, as B4 isomerizes to B2 and cC. The ratio of the two phases due to these two remains constant, however, since the equilibrium between them is always maintained. Extrapolation of the set of traces of Figure 12 to zero time of standing of the neutral solution gives the relative amount of the three neutral products obtained on initial hydrolysis, that is, the kinetic product mixture. For Figure 12 this extrapolation produces the estimate that at such time 50–55% B4 is present. It can be recalled that according to our kinetic analysis two-thirds B4 is formed on hydration of F^+ with a water molecule, so that the two estimates are not too different. Moreover, at pH 8.3 a significant fraction (about 20–25%) of the disappearance of F^+ involves its reaction with OH^-, and this combination, as we will show below, produces less B4 as kinetic product. Unfortunately, this sort of analysis cannot be carried out for solutions of lower pH where hydration involves only water, since the isomerization then becomes too rapid and extrapolation is very difficult.

In order to obtain the remaining rate constants for F^+–pseudobase interconversion the ratio of B4:B2 produced as kinetic products of the reaction of OH^- and F^+ is needed. This quantity is obtained by applying the extrapolation procedure described in the last paragraph to a strongly basic solution of F^+, that is, where hydrolysis involves only OH^-. With all three ions this analysis shows that one-third of the initial product mixture in base is B4. This result produces

$$k_4^{OH}/k_1^{OH} = 0.5 \tag{29}$$

Since the sum of k_4^{OH} and k_1^{OH} is known (eq 25), the individual values can be calculated. These can then be combined[26] with the known acidity constants to give values of k_{-4}^{w} and k_{-1}^{w}.

There is one piece of information which has not been used in any derivation, and which provides now a check on the consistency of the analysis. That is the rate expression for k_{obsd}^{w}(isom), eq 28, for which all the rate constants have now been independently evaluated. These produce a calculated value of k_{obsd}^{w}(isom) of 0.0012 s^{-1}, while the observed value is 0.0015 s^{-1}. Considering the number of observed parameters with their inherent errors which are involved in obtaining the derived rate constants, the agreement must be regarded as satisfactory.

One final observation to be explained is the complex kinetics of F^+ disappearance found at certain acidities (Figure 8). To account for this we recognize that what is involved in these solutions is the kinetic system

$$\mathrm{B4} \underset{\nu_{-4}}{\overset{\nu_4}{\rightleftharpoons}} \mathrm{F^+} \xrightarrow{\nu_1} \mathrm{B2} \tag{30}$$

where ν_1, ν_4, and ν_{-4} are first-order rate constants at a given pH. We have solved the differential equations for this system,[45] to obtain a solution expressing the concentration of F^+ as a function of time.[46] Substitution into this equation of the values at the appropriate pH of ν_1, etc., calculated as $k_1^{w} + k_1^{OH}[OH^-]$, etc., reproduces the observed traces (Figure 7).

These complex traces have their origin in the fact that the F^+–B4 equilibrium does not completely favor B4. In simple terms, a rapid decrease in F^+ is observed as this equilibrium is established, followed by a slower decrease as the system siphons toward B2. The first-order kinetics observed at higher pH arise when the F^+–B4 equilibrium does strongly favor B4. Substitution into the exact equation[46] of the condition that $\nu_4 \ll \nu_{-4}$ collapses that equation to a first-order expression with $k_{obsd} = \nu_1 + \nu_4$, as in eq 23.

***cis*-Chalcone–*trans*-Chalcone Interconversion.** Equation 31 presents our model to account for the kinetics of formation of the final product in most solutions, the *trans*-2-hydroxychalcone. In

$$\begin{tikzcd}
\mathrm{F^+} \arrow[r, shift left, "K_1^a"] & \mathrm{B2} \arrow[l, shift left] \arrow[r, shift left, "K_2"] & \mathrm{cC} \arrow[l, shift left] \arrow[r, "k_3^w"] \arrow[d, shift left] & \mathrm{tC} \\
 & & \mathrm{cC^-} \arrow[u, shift left, "K_{cC}^a"] \arrow[r, "k_{3'}^w"] & \mathrm{tC^-}
\end{tikzcd} \tag{31}$$

this mechanism[47] *trans*-chalcone or its anion arise from the isomerization of *cis*-chalcone or its anion, respectively, the latter two species being in equilibrium with each other and with F^+ and B2. That is, the various transformations involving F^+, B2, cC, and cC^- (and also B4) are complete and these species are at equilibrium before the cis–trans isomerization occurs. Furthermore, the rate at which these species equilibrate is more rapid than the rate of cis–trans isomerization so that the equilibrium is maintained. Where the concentration of F^+ in the equilibrium is negligible (pH >6), eq 32 can be derived for the observed rate constants for tC formation.

$$k_{obsd}(\mathrm{tC}) = \frac{k_{3'}^{w}\left(\dfrac{K_2}{1+K_2}\right)K_{cC}^{a} + \left(\dfrac{K_2}{1+K_2}\right)k_3^{w}[H^+]}{\left(\dfrac{K_2}{1+K_2}\right)K_{cC}^{a} + [H^+]} \tag{32}$$

This has the same form as the observed equation, eq 3, with the empirical constants being given by

$$k_{obsd}^{base}(\mathrm{tC}) = k_{3'}^{w} \tag{33}$$

$$k_{obsd}^{neut}(\mathrm{tC}) = \left(\frac{K_2}{1+K_2}\right)k_3^{w} \tag{34}$$

$$K_{obsd}^{a}(\mathrm{base}) = \left(\frac{K_2}{1+K_2}\right)K_{cC}^{a} \tag{35}$$

Equation 35 is the same as eq 16, which refers to the acidity constant obtained spectroscopically. The two acidity constants refer to the same equilibrium and should therefore be the same. It can be noted that in strong base the cis–trans isomerization occurs via the *cis*-chalcone anion, which is the predominant species of the equilibrium with cC and B2. The observed rate constant is pH independent, and is equal to $k_{3'}^{w}$. As the pH is decreased there is a changeover region, followed in neutral solutions by a second pH-independent region, representing the isomerization of the neutral *cis*-chalcone. The observed rate constant in this region is given by eq 34, and is less than k_3^{w} because the concentration of the isomerizing species cC is lowered by its being in equilibrium with the unreactive B2. In more acidic solutions isomerization continues to occur via the neutral *cis*-chalcone, but the rate decreases further from k_3^{w} as the acid further decreases the concentration of cC by converting it reversibly to F^+. The observed rate constant (pH <6) is given by eq 36, which can be compared with the observed eq 4.

$$k_{obsd}(\mathrm{tC}) = \left(\frac{K_2}{1+K_2}\right)k_3^{w}\left(\frac{K_1^{a}(1+K_2)}{K_1^{a}(1+K_2) + [H^+]}\right) \tag{36}$$

It can be noted that the limiting rate constant in acids where $[F^+]$ dominates [B2] and [cC] is given by

$$k_{obsd}(\mathrm{tC}) = k_3^{w}K_1^{a}K_2/[H^+] \tag{37}$$

This shows an inverse dependency on the $[H^+]$ concentration.

The last experiment described in the Results section places a lower limit of 5000 on the equilibrium constant for the reversible isomerization of cC^- and tC^-. If the assumption is made that the acidity constants of the two chalcones are similar, the same lower

---

(45) Ritchie, C. D. "Physical Organic Chemistry. The Fundamental Concepts"; Marcel Dekker: New York, 1975; pp 7–30.

(46) $[F^+]/[F^+]_0 = \{(\nu_{-4} + \lambda_2)/(\lambda_2 - \lambda_1)\}e^{\lambda_2 t} - \{(\nu_{-4} + \lambda_1)/(\lambda_2 - \lambda_1)\}e^{\lambda_1 t}$, where λ_1 and $\lambda_2 = -(1/2)\{(\nu_1 + \nu_4 + \nu_{-4}) \pm [(\nu_1 + \nu_4 + \nu_{-4})^2 - 4\nu_{-4}\nu_1]^{1/2}\}$.

(47) One can write other mechanisms. For example, a Michael addition of OH^- to the neutral *cis*-chalcone, followed by single bond rotation, and loss of OH^- is kinetically equivalent to isomerization of cC^-. We are currently carrying out further investigations of the isomerization and will defer a detailed discussion of the mechanism until that is completed.

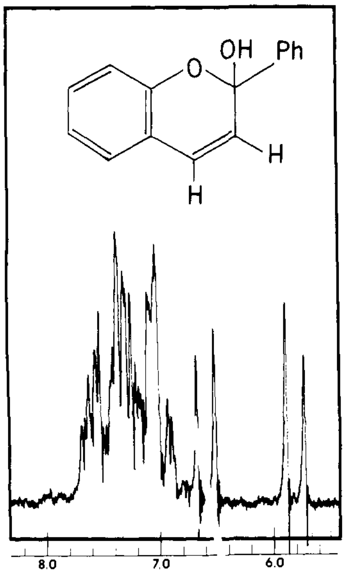
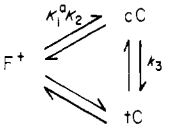


Figure 13. NMR spectrum of the pseudobase B2 of the flavylium ion.

limit can be placed on K_3, the equilibrium constant for isomerization of the two neutral chalcones. Referring to the scheme of eq 38, the equilibrium constant for interconversion of F^+ and tC



(38)

can also now be provided with a limit, $pK_a(F^+ \rightleftharpoons tC) < 2$, this limit applying to the 4′-methoxy system, and being even lower with the other two. The point of this analysis is that it demonstrates that the flavylium ions of this study are not thermodynamically stable toward *trans*-chalcone even in quite acidic solutions, perhaps even the stock solution of 0.1 M HCl. This would explain the failure of *trans*-chalcone to convert to flavylium ion in 0.1 M HCl, while such a reaction does proceed in more concentrated acids where the $F^+ \rightleftharpoons$ tC equilibrium would certainly favor F^+. The stability of the flavylium ion in the pH 1–2 solutions then becomes a kinetic stability, attributable to the low equilibrium concentration of the isomerization species cC. For example, according to eq 36, the half-life for the formation of *trans*-chalcone from the 4′-methoxyflavylium ion at 25 °C[48] in 0.1 M HCl is 815 days.

Summary

Summarized below are the patterns of behavior exhibited by the 4′-methoxyflavylium ion at four selected pH values, chosen to illustrate the basic features of the system.

(**a**) **pH 1.0.** The flavylium ion is stable. Whether it is thermodynamically stable toward *trans*-chalcone or simply kinetically stable requires further experiment.

(**b**) **pH 4.29.** An equilibrium is rapidly established (half-life ≈ 2 s, although the kinetics are likely complex), consisting of 50% F^+, 33.2% B2, 0.3% B4, and 16.5% cC. A slower reaction follows (half-life 19.7 h), resulting in complete conversion to tC.

(**c**) **pH 8.0.** The flavylium ion reacts mainly with solvent water (half-life = 0.44 s), producing 64% B4, 24% B2, and 12% cC, the last two being in equilibrium with each other (half-life of equilibration = 7×10^{-5} s at pH 8). This is followed by a reaction (half-life 66 s) in which B4, a product of kinetic control of the initial neutralization of F^+, is converted via F^+ to B2 and cC, the product mixture at equilibrium consisting of 66.3% B2, 33.1% cC, and 0.6% B4. A slower reaction (half-life 9.9 h) then occurs, resulting in complete conversion to tC.

(**d**) **pH 12.0.** The flavylium ion reacts with hydroxide ion (half-life = 3.8×10^{-4} s), producing a mixture of B4[49] and cC^- in a ratio of 1:2. The latter ion is formed rapidly from the initially produced B2, the equilibrium between B2 and *cis*-chalcone being displaced toward the latter because of its ionization. B4 is then converted (half-life = 7.7 min) via F^+ and B2 to cC^-, producing a solution containing >99% of this anion. This undergoes a slow isomerization (half-life = 4.2 h), producing tC^-.[50]

It can be seen that *trans*-chalcone, or its anion, is the ultimate product at all pH, except perhaps in the most acidic solutions. The upper limit on the concentration of *cis*-chalcone, or its anion, which is present in equilibrium with this when equilibrium is established is 0.02%.

Experimental Section

Equipment. NMR spectra were obtained on a Varian T60 NMR spectrophotometer. Spectral measurements and conventional spectroscopic kinetic measurements were made on either a Unican SP 1800 or Cary Model 118C spectrophotometer. Both instruments are equipped with jacketed multiple cell blocks maintained at 25.0 ± 0.01 °C by external water baths, and automatic sample changing and recording facilities. Stopped-flow kinetic measurements were made on a Durrum-Gibson stopped-flow spectrophotometer operating at 25.0 ± 0.1 °C. The photomultiplier output of this spectrophotometer was converted to digital form and fed directly to a Tektronix 4051 minicomputer equipped with graphics display, where it was analyzed. Our setup records 118 absorbance readings in a given kinetic run. The optical density resolution is 2^{14} and the time period can be varied from 0.002 to 100 s.

Materials. The perchlorate salts of the three flavylium ions were prepared by the acid condensation of salicylaldehyde and the appropriate acetophenone.[51,52] 2-Hydroxychalcone and 2-hydroxy-4′-methoxychalcone were prepared by base condensation of salicylaldehyde and the appropriate acetophenone.[53] All other materials were best available commercial grades.

Methods. The flavylium perchlorates were handled by preparing a stock 1–2 × 10^{-4} M solution of the ion in 0.1 M HCl, and neutralizing or diluting this solution to obtain solutions for spectral or kinetic studies.

Stopped-flow kinetic studies involving neutral and base solutions (disappearance of flavylium ion, k_{obsd}(dis)) were carried out by placing the stock acid solution in one syringe of the stopped-flow apparatus and an appropriate buffer containing 0.1 M NaOH in the other syringe. The wavelength maximum of the flavylium ion was monitored (397 nm, unsubstituted; 411 nm, 4′-methyl; 440 nm, 4′-methoxy). Rate constants, where the disappearance is first order, were obtained as slopes of plots of ln $(A - A_\infty)$ vs. time.

Stopped-flow kinetic studies involving reacidification of neutral or base solutions were carried out by adding the stock acid solution to a very dilute buffer or base solution also containing 0.1 M NaOH, and placing this in one syringe of the stopped-flow apparatus, with an appropriate HCl solution in the other syringe. The λ_{max} of the flavylium ion was monitored. Rate constants for the fast phase ($k_{obsd}(F^+$-fast)) were obtained by using the Guggenheim method. Rate constants for the slow phase ($k_{obsd}(F^+$-slow)) were obtained as slopes of plots on ln $(A_\infty - A)$ vs. time.

Rate constants for the intermediate kinetic phase (k_{obsd}(isom)) were measured on the Cary 118 spectrophotometer at 290–300 nm, using the Guggenheim method.

Rate constants for the formation of *trans*-chalcone (k_{obsd}(tC)) were measured following the absorbance increase at 320–350 (pH <9.5) or 430–440 nm (pH >9.5) and are based on the infinity method or the Guggenheim method, the latter being used for very slow runs.

All first-order kinetic plots were accurately linear over several half-lives with correlation coefficients of 0.999 or greater (usually 0.9999). Rate constants were obtained as the average of two to three kinetic runs, using conventional spectroscopy and five to eight kinetic runs and stopped-flow spectroscopy. The ionic strength of all solutions was maintained constant at 0.1 by the addition of NaCl. The pH of all solutions was measured on a Radiometer pH meter equipped with combination electrode. With the exception of the stopped-flow studies, experiments were

---

[48] We are currently carrying out experiments by using elevated temperatures to set up the $F^+ \rightleftharpoons$ tC equilibrium.

[49] In very concentrated base solutions (pH >12) B4 is probably also significantly ionized.

[50] A referee also suggested the possibility of the formation of 2′-hydroxy 4-substituted chalcones. Such species could form from the B4 pseudobase, followed by loss or transfer of H_4. The spectral considerations would seem, however, to rule these out.

[51] Kokkinos, K.; Wizinger, R. *Helv. Chim. Acta* **1973**, *56*, 983.

[52] Michaeldis, C.; Wizinger, R. *Helv. Chim. Acta* **1951**, *34*, 1761.

[53] Herstein, F.; von Kostanecki, S. *Ber. Dtsch. Chem. Ges.* **1899**, *32*, 318.

carried out as much as possible in the dark. This was particularly important in studying the cis → trans isomerization. This reaction, and its reverse, appear to be affected by prolonged exposure to light.

Buffer catalysis was observed for several of the processes (see text) and in these cases rate constants were extrapolated to zero buffer concentration. The total concentration of the buffers used in our studies ranged from 0.1 to 0.005 M. With the exception of the B2 $\rightleftarrows$ cC equilibration (eq 17), the contribution of the buffer to the overall rate was small, the buffer in the most concentrated solution accounting for no more than 25% of the rate.

Acknowledgments. The financial support of the Natural Sciences and Engineering Research Council of Canada and the Research Corporation is gratefully acknowledged.

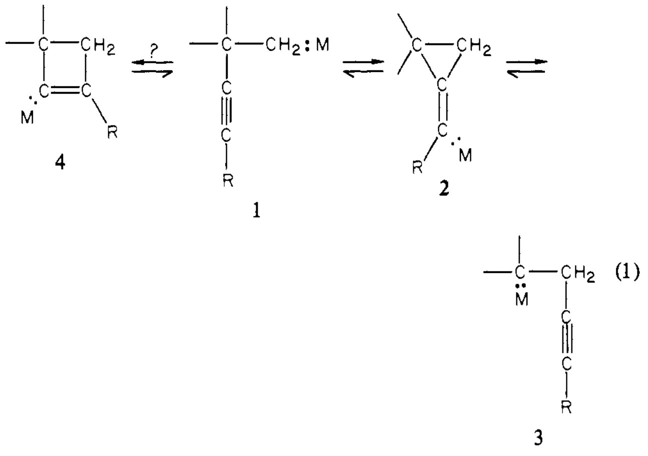
Carbanions 20. Rearrangements in Reactions of 5-Chloro-4,4-diphenyl-2-pentyne, 1-Chloro-5,5-dimethyl-2,2-diphenyl-3-hexyne, and 5,5-Dimethyl-2,2-diphenyl-3-hexyne with Alkali Metals. Cation Effects on Migratory Aptitudes of Groups

Erling Grovenstein, Jr.,* Kuen-Wai Chiu, and Bhalchandra B. Patil

Contribution from the School of Chemistry, Georgia Institute of Technology, Atlanta, Georgia 30332. Received March 17, 1980

Abstract: Lithium reacts with 5-chloro-4,4-diphenyl-2-pentyne (**7a**) and with 1-chloro-5,5-dimethyl-2,2-diphenyl-3-hexyne (**7b**) in THF at −75 °C to give 2,2-diphenyl-3-pentynyllithium (**14a**) and 5,5-dimethyl-2,2-diphenyl-3-hexynyllithium (**14b**), respectively; when the THF solutions of these organolithium reagents are warmed to 0 °C, [1,2] migration of the acetylenic group of each occurs to produce 1,1-diphenyl-3-pentynyllithium (**16a**) and 5,5-dimethyl-1,1-diphenyl-3-hexynyllithium (**16b**), respectively, as deduced from the products of carbonation. Reaction of the organolithium compound **14a** with cesium *tert*-butoxide in THF at −75 °C gives a propargylic anion of unrearranged carbon skeleton from metalation of the methyl group; similar treatment of **14b** gives an organoalkali product derived from metalation of the ortho position of a neighboring phenyl group again without rearrangement of carbon skeleton. Reaction of the chloride **7b** with Cs–K–Na alloy in THF at −75 °C gives, evidently first, 5,5-dimethyl-2,2-diphenyl-3-hexynylcesium (**32**), which then undergoes [1,2] migration of phenyl to give 1-benzyl-4,4-dimethyl-1-phenyl-2-pentynylcesium (**27**). Much of **27** is metalated by **32** to give the dianion **28** and 5,5-dimethyl-2,2-diphenyl-3-hexyne (**18**). Hydrocarbon **18**, as confirmed by independent experiments, reacts with Cs–K–Na alloy to give a dianion **31** of rearranged carbon skeleton along with biphenyl. These products from the reaction with Cs–K–Na alloy were characterized as the hydrocarbons obtained by protonation and deuteration. The hydrocarbon from **31** is the novel compound 5,5-dimethyl-2-(1-phenyl-2,5-cyclohexadienyl)-2,3-hexadiene (**26**). An explanation is proposed for the change in relative migratory aptitude of acetylenic and phenyl groups with change of alkali metal cation from lithium to cesium. Also the protonation and alkylation of propargylic anions are discussed.

Vinyl groups have been observed to undergo [1,2] sigmatropic rearrangements in organoalkali compounds[1] and in Grignard reagents[2] while phenyl groups have been reported to migrate in organoalkali compounds but, so far, not in Grignard reagents[3] in ethereal solvents.[4] Some time ago one of us predicted[5] that a [1,2] shift of an acetylenic group in a carbanion was a likely possibility, e.g., **1** → **3** (M = alkali metal). By analogy with rearrangements of vinyl and phenyl groups in organometallic compounds, rearrangement of an acetylenic group would be expected to proceed by way of a cyclic intermediate or transition state such as **2** formed by addition of the organometallic moiety to the triple bond.



While a number of examples[2,6-8] of intramolecular cyclization of acetylenic Grignard and lithium reagents to give five- or six-membered rings are now known, only one example[9] of a [1,2] shift of an acetylenic group in a Grignard reagent (none in an orga-

---

[1] For reviews see: (a) Grovenstein, E. *Adv. Organomet. Chem.* **1977**, *16*, 167–210. (b) *Angew. Chem., Int. Ed. Engl.* **1978**, *17*, 313–332.

[2] For reviews see: (a) Hill, E. A. *J. Organomet. Chem.* **1975**, *91*, 123–271. (b) *Adv. Organomet. Chem.* **1977**, *16*, 131–165.

[3] (a) Zimmerman, H. E.; Zweig, A. *J. Am. Chem. Soc.* **1961**, *83*, 1196–1213. (b) Grovenstein, E.; Cottingham, A. B.; Gelbaum, L. T. *J. Org. Chem.* **1978**, *43*, 3332–3334.

[4] Wittig ether rearrangement with phenyl migration has been observed in an organomagnesium compound in the good ionizing solvent hexamethylphosphoramide evidently by way of a solvent-separated ion pair (Ebel, H. F.; Dörr, V.; Wagner, B. O. *Angew. Chem., Int. Ed. Engl.* **1970**, *9*, 163).

[5] Grovenstein, E.; Wentworth, G. *J. Am. Chem. Soc.* **1967**, *89*, 2348–2356.

[6] (a) Kandil, S. A.; Dessy, R. E. *J. Am. Chem. Soc.* **1966**, *88*, 3027–3034. (b) Richey, H. G.; Rothman, A. M. *Tetrahedron Lett.* **1968**, 1457–1460. (c) Hill, E. A.; Theissen, R. J.; Doughty, A.; Miller, R. *J. Org. Chem.* **1969**, *34*, 3681–3684.

[7] (a) Crandall, J. K.; Keyton, D. J. *Tetrahedron Lett.* **1969**, 1653–1656. (b) Moore, W. M.; Salajegheh, A.; Peters, D. G. *J. Am. Chem. Soc.* **1975**, *97*, 4954–4960.

[8] Courtois, G.; Masson, A.; Migniac, L. *C. R. Acad. Sci., Ser. C* **1978**, *286*, 265–268.

[9] Richey, H. G.; Rothman, A. M., personal communication to E. A. Hill quoted in ref 2a, p 150.

0002-7863/80/1502-5848$01.00/0