After crystallization from benzene–petroleum ether (40–70 °C), the product **8** had the following: mp 132–138 °C dec; $[\alpha]_D$ −69.9° (*c* 1.0); IR ν_{max} 3600, 3420 (OH), 1720 (C=O) cm^{-1}; ^1H NMR 1.00 (3 H, s, C_{10}-CH_3), 1.12 (3 H, s, C_{13}-CH_3), 3.42 (3 H, s, OCH_3), 3.60 (1 H, br signal, CHOH), 4.15 (1 H, nearly four broad signals, $J \simeq 12$, 8 Hz, $CHOCH_3$), 5.35 (1 H, br signal, CH=); mass spectrum, *m/e* 332 (M^+, base), 271, 239, 201. Anal. Calcd for $C_{21}H_{32}O_3$: C, 75.86; H, 9.70. Found: C, 75.70; H, 9.85.

After crystallization from CH_3OH, the product **9** had the following: mp 207–215 °C dec; $[\alpha]_D$ + 103.1° (*c* 0.8, dioxane); IR ν_{max} 3600, 3300 (OH), 1720 (C=O) cm^{-1}; ^1H NMR (dioxane-d_8, sparingly soluble) 1.10 (3 H, s, C_{13}-CH_3), 3.30 (3 H, s, OCH_3), 4.05 (1 H, br signal, $CHOCH_3$), 6.45 (2 H, 2 br signals) and 7.08 (1 H, 2 br signals) (aromatic protons); mass spectrum, *m/e* 314 (M^+, base), 213, 160. Anal. Calcd for $C_{20}H_{26}O_3$: C, 76.40; H, 8.34. Found: C, 76.35; H, 8.50.

Preparation of 17-Methylene-1,3,5-estratrien-3-yl Acetate (5) and Its 3-Hydroxy Derivative. Compound **5** was prepared from 3-hydroxy-1,3,5-estratrien-17-one according to the procedure described for other 17-ketones,[4] yield 80%.

After crystallization from CH_3OH, the product **5** had the following: mp 80–82 °C; $[\alpha]_D$ +51.8° (*c* 1.5); IR ν_{max} 1760 (OC-OCH_3), 880 (C=CH_2) cm^{-1}; ^1H NMR 0.82 (3 H, s, C_{13}-CH_3), 2.22 (3 H, s, CH_3C=O), 4.69 (2 H, br signal, C=CH_2), 6.84 (2 H, 2 br signals) and 7.29 (1 H, 2 br signals) (aromatic protons); mass spectrum, *m/e* 310 (M^+), 268 (base), 269, 160, 133. Anal. Calcd for $C_{21}H_{26}O_2$: C, 81.25; H, 8.44. Found: C, 81.50; H, 8.64.

The 3-hydroxy derivative of **5** (17-methylene-1,3,5-estratrien-3-ol) was obtained by treating **5** with methanolic KOH (5–7% w/v) solution, following the same procedure used for the hydrolysis of **2**, **4**, and **6**. The product was then crystallized from benzene–petroleum ether (40–70 °C) and had the following: mp 134–137 °C; $[\alpha]_D$ 49.9° (*c* 0.7); IR ν_{max} 3490, 3310 (OH), 8.75 (C=CH_2) cm^{-1}; ^1H NMR 0.84 (3 H, s, C_{13}-CH_3), 4.70 and 4.95 (2 H, 2 br signals, C=CH_2), 6.63 (2 H, 2 br signals) and 7.17 (1 H, 2 br signals) (aromatic protons); mass spectrum, *m/e* 260 (M^+, base), 160, 133. Anal. Calcd for $C_{19}H_{24}O$: C, 85.02; H, 9.01. Found: C, 84.80; H, 9.01.

Registry No. **1**, 1164-94-9; **2**, 77257-04-6; **3**, 853-22-5; **4**, 77257-05-7; **5**, 77257-06-8; **6**, 77257-07-9; **7**, 77257-08-0; **8**, 77257-09-1; **9**, 77257-10-4; 3-hydroxy-1,3,5-estratrien-17-one, 53-16-7; 17-methylene-1,3,5-estratrien-3-ol, 34111-53-0; TTN, 13746-98-0.

Analysis of Kinetic Isotope Effects on Complex Reactions Utilizing the Concept of the Virtual Transition State

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Kinetic isotope effects are observed when reaction rates are compared for molecules differing only in isotopic composition and originate from the different sensitivities of the reactant and transition state vibrational force fields to the isotopic substitution.[1,2] Interpretation of an observed isotope effect ultimately results in the assignment of structures, having vibrational modes of the appropriate isotopic sensitivity, to the transition state and reactant. Problems in interpretation may arise for reactions having more than a single transition state. Reactions having several transition states, "complex" reactions, are those which proceed through at least one intermediate and/or by way of at least two pathways. For such reactions in which more than a single transition state is kinetically significant, any experiment designed to probe the structure of the rate-limiting transition state will yield information not about a single, real transition state, but rather about a "virtual" transition state whose structure is a weighted average of structures of the several rate-determining, real transition states.[3] Here the problem becomes one of dissecting structures of real transition states out of the composite structure experimentally accessible. If the experimental probe of transition-state structure is the kinetic isotope effect, then this problem can, in principle, be solved by first determining the rate constants for all terms of the rate law for both isotopically labeled substrates and then dividing corresponding rate constants to produce isotope effects on the inidividual reaction steps. Such a procedure is, in fact, of limited applicability. Very high precision in the determination of reaction rates is required for the exact determination of individual rate constants and will be quite difficult to achieve for most complex reactions of interest. Even if precise data can be obtained, fitting of the data to a rate law by curve-fitting procedures to get rate constants for all terms of the rate law may prove mathematically or computationally impossible.[4] Under certain circumstances, however, a procedure not requiring the precise determination of the individual terms of a rate law is available for the quantitative analysis of kinetic isotope effect data.

We begin with the idea that the observed kinetic isotope effect on a complex reaction can be expressed as a weighted average of isotope effects on the individual reaction steps (k_m/k_m^*). The weighting factors (C_m) are the contribu-

$$k_0/k_0^* = C_1(k_1/k_1^*) + C_2(k_2/k_2^*) + ...C_m(k_m/k_m^*) \quad (1)$$

tions from the transition state of each step to determining the structure of the virtual transition state. Note that the rate constant, k_m, for the *m*th step reflects the free-energy difference between the ground state and the *m*th transition state, TS-*m*; that is, k_m is equal to the product of equilibrium constants for all steps leading to TS-*m* multiplied by the rate constant for reaction over TS-*m*.[5]

The weighting factors, C_m, can be calculated in a straightforward way. For a reaction having consecutive transition states, it can be shown that

$$C_m = (e^{\Delta G_m\dagger/RT}/e^{\Delta G\dagger_0/RT}) \quad (2)$$

or

$$C_m = k_0/k_m \quad (3)$$

where the apparent activation energy ($\Delta G_0^\dagger$) is the free-energy difference between the reactants and the virtual transition state. Similarly, for a reaction proceeding through competitive, parallel pathways

$$C_m = (e^{-\Delta G_m\dagger/RT}/e^{-\Delta G_0\dagger/RT}) \quad (4)$$

or

$$C_m = k_m/k_0 \quad (5)$$

Note that in this case the *lowest* free-energy transition

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(5) This treatment, as derived here, applies only to those reactions having intermediates of insignificant lifetime. Similar, but more complex, equations can be derived which apply to reactions having intermediates of appreciable lifetime.

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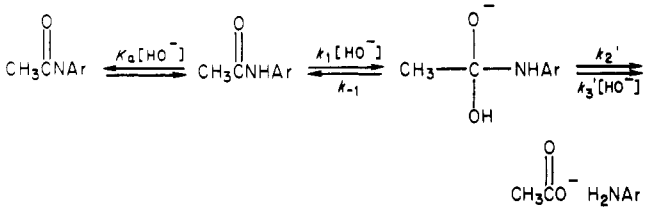
Scheme I



state contributes the most to determining the reaction rate, while for reactions having serial transition states, the *highest* free-energy transition state is the heaviest contributor.

Inspection of eq 1 reveals that even if the isotope effect (k_0/k_0^*) and transition-state contributions are known for a reaction it is still not possible to calculate the isotope effects on the individual reaction processes; we are faced with the algebraic problem of too many unknowns for too few simultaneous equations. A special case and the one in which we are interested occurs when these transition-state contributions are a function of some reaction variable, X. It then becomes possible to measure isotope effects and calculate values of C_m at many values of X and thus be able to solve for the isotope effects on discrete reaction processes.

We see then that for multistep, multipath reactions where the structure of the virtual transition state can be fine-tuned by adjustment of a reaction variable, eq 1, 3, and 5, taken together, are a recipe for calculating the isotope effects on the individual reaction steps.

An example of the application of these principles is found in a recent study from this laboratory of the basic hydrolysis of *p*-nitroacetanilide (PNAA) where the kinetic β-deuterium isotope effect (PNAA vs. acetyl-d_3-PNAA) was determined as a function of hydroxide concentration.[6] Relevant data are collected in Table I. The mechanism for the basic hydrolysis of acetanilides has been determined in some detail[7] and is summarized for *p*-nitroacetanilide in Scheme I.

The rate law for this mechanism is shown by

$$k_0 = \frac{k_1[HO^-](k_2 + k_3[HO^-])}{k_1 + k_2 + k_3[HO^-]} \quad (6)$$

where

$$k_0 = k_{obsd}(1 + K_a[HO^-]) \quad (7)$$

$$k_2 = \left(\frac{k_1}{k_{-1}}\right)k_2' \quad (8)$$

$$k_3 = \left(\frac{k_1}{k_{-1}}\right)k_3' \quad (9)$$

The reciprocal of the rate law, eq 10, is an expression which

$$\frac{1}{k_0} = \frac{1}{k_1[HO^-]} + \frac{1}{k_2[HO^-] + k_3[HO^-]^2} \quad (10)$$

relates the observed rate to terms expressing the resistance encountered when passing over the energy barriers of this reaction. The first term on the right hand side of eq 10 is a measure of the resistance reactants encounter when passing through the transition state of k_1, while the second term measures the resistance encountered when passing through a second, consecutive transition state, either that of k_2 or k_3 or some composite of these transition states.

Table I. Kinetic Data for the Basic Hydrolysis of *p*-Nitroacetanilide

$[HO^-]$,[a] M	$10^5 k_0$,[b] s^{-1}	k_0^H/k_0^D ±0.01	transition-state contributions[c] C_1	C_2	C_3
2.31	988	0.927	0.95	0.00	0.05
1.00	411	0.946	0.90	0.00	0.10
0.208	61.7	0.971	0.65	0.01	0.34
0.075	12.9	0.973	0.41	0.02	0.57
0.028	2.69	0.979	0.21	0.05	0.74
0.010	0.394	0.977	0.10	0.13	0.78
0.005	0.135	0.964	0.05	0.24	0.71
0.0016	0.0286	0.949	0.00	0.50	0.50

[a] Sodium hydroxide concentrations were determined by titration against sodium hydrogen phthalate with a phenolphthalein end point. [b] First-order rate constants for the basic hydrolysis of *p*-nitroacetanilide determined at 30.17 ± 0.05 °C and μ = 0.50 (NaCl) for hydroxide concentrations less than 0.50 M. For reactions catalyzed by hydroxide at concentrations of 2.31, 1.00, and 0.208 M, data (OD_{410}) were collected for at least three half-lives and pseudo-first order constants determined by nonlinear least-squares analysis of the time course. For less basic hydroxide solutions data were collected for less than 1% conversion and rate constants determined by linear least-squares analysis of the time course. [c] Calculated from eq 6, 12, 16, and 18 with rate constants $k_1 = 4.5 \times 10^{-3}$ M^{-1} s^{-1}, $k_2 = 9.2 \times 10^{-5}$ M^{-1} s^{-1}, $k_3 = 4.6 \times 10^{-2}$ M^{-2} s^{-1}.

The form of the rate law expressed in eq 10 is especially helpful for the calculation of transition-state contributions for this reaction: the two terms on the right hand side of eq 10 are related in a simple way to the transition-state contributions, C_1 and $C_{[2,3]}$ of two serial transition states, TS-1 and TS-[2,3].

For TS-1

$$C_1 = (1/k_1[HO^-])/(1/k_0) \quad (11)$$

$$C_1 = k_0/k_1[HO^-] \quad (12)$$

and for TS-[2,3]

$$C_{[2,3]} = [1/(k_2[HO^-] + k_3[HO^-]^2)]/(1/k_0) \quad (13)$$

$$C_{[2,3]} = k_0/(k_2[HO^-] + k_3[HO^-]^2) \quad (14)$$

C_1 and $C_{[2,3]}$ both have the general form of eq 3: transition-state contributions from serial transition states.

TS-[2,3] is of course a composite of transition states, TS-2 and TS-3, for two competitive, parallel paths, k_2 and k_3. Here

$$C_2 = C_{[2,3]}\left(\frac{1/(k_2[HO^-] + k_3[HO^-]^2)}{1/k_2[HO^-]}\right) \quad (15)$$

$$C_2 = C_{[2,3]}\left(\frac{k_2[HO^-]}{k_2[HO^-] + k_3[HO^-]^2}\right) \quad (16)$$

and

$$C_3 = C_{[2,3]}\left(\frac{1/(k_2[HO^-] + k_3[HO^-]^2)}{1/k_3[HO^-]^2}\right) \quad (17)$$

$$C_3 = C_{[2,3]}\left(\frac{k_3[HO^-]^2}{k_2[HO^-] + k_3[HO^-]^2}\right) \quad (18)$$

C_2 and C_3 have the form of eq 5: transition-state contri-

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Table II. Kinetic β-Deuterium Isotope Effects on the Reaction Steps for the Basic Hydrolysis of *p*-Nitroacetanilide

rate constant[a]	k_n^H/k_n^D [b]
k_1	0.938 ± 0.007
k_2	0.902 ± 0.020
k_3	1.000 ± 0.010

[a] See text for definition of rate constants. [b] Best fit values from a nonlinear least-squares fit of the k_0^H/k_0^D vs. C_m data (Table I) to eq 1.

Table III. Calculated Isotope Effects for Values of k_3/k_2

k_3/k_2, M^{-1}	isotope effects[a]		
	k_1	k_2	k_3
200	0.936	0.927	1.012
400	0.937	0.910	1.003
500	0.938	0.902	1.000
600	0.938	0.895	0.999
800	0.939	0.880	0.997

[a] Calculated from nonlinear least-squares fit of the experimental data of Table I to eq 19 using the rate constants $k_1 = 4.5 \times 10^{-3}$ M^{-1} s^{-1} and $k_3 = 4.6 \times 10^{-2}$ M^{-1} s^{-1}.

butions from transition states on parallel paths.

Finally, then, we see that the observed isotope effect on PNAA hydrolysis is equal to eq 19.

$$\frac{k_0^H}{k_0^D} = \frac{k_0}{k_1[HO^-]}\left(\frac{k_1^H}{k_1^D}\right) + \frac{C_{[2,3]}}{k_2[HO^-] + k_3[HO^-]^2}\left(k_2[HO^-]\frac{k_2^H}{k_2^D} + k_3[HO^-]^2\frac{k_3^H}{k_3^D}\right) \quad (19)$$

Nonlinear least-squares fit of the k_0^H/k_0^D vs. C_m data (Table I) to eq 1 allows the determination of the isotope effects of k_1, k_2, and k_3. These values are collected in Table II. (Mechanistic interpretation of the isotope effect will be presented elsewhere.[6]) It should be noted that although values for three rate constants are needed for calculation of transition-state contributions, one of these constants, k_2, could not be determined.[6] In the present analysis k_2 was assigned what we believe to be a reasonable value of 9.2×10^{-5} M^{-1} s^{-1} ($k_3/k_2 = 500$ M^{-1}). Of course it is of interest to know how sensitive the calculated isotope effects are to k_2, and in Table III are collected the calculated isotope effects for several values of k_2. Clearly, the isotope effects, especially on k_1 and k_3, are insensitive to k_2 and, indeed, at the two extremes of k_2 which mark limits of reasonable values the isotope effects generated lead one to identical qualitative mechanistic interpretations.[6]

The procedure outlined here for the analysis of observed isotope effects on rates of complex reactions is a general one and can be applied to kinetic isotope effect data for any reaction having a virtual transition state whose structure can be varied by adjustment of some reaction parameter which influences the relative free-energy barriers of the individual reaction steps. The utility of this procedure lies in its ability to extract isotope effects for individual reaction processes from complex, observed isotope effects without precise knowledge of rate constants which may frequently be difficult if not impossible to obtain.

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Registry No. *p*-Nitroacetanilide, 104-04-1.

Isolation, Identification, and Synthesis of Compounds Cosynthesized in the Preparation of Phencyclidine[1]

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The addition of 1-piperidinocyclohexanecarbonitrile (PCC, **1**) to phenylmagnesium bromide produces 1-(1-phenylcyclohexyl)piperidine (PCP, **2**),[2] a dissociative analgesic which has become a drug of abuse[3] and is primarily self-administered by smoking PCP "doped" cigarettes.[4] Preparatory to the study of the pyrolysis products of **2** under simulated smoking conditions, we examined those compounds containing the phenyl and piperidyl groups cosynthesized in the Kalir synthesis[2] of this compound since high-pressure liquid chromatography (HPLC)[5] indicated at least 13 additional nitrogen-containing compounds were present in the crude basic fraction of this preparation. The isolation, identification, and bioassay of these cosynthetics were undertaken and the results are reported herein.

Open column chromatography on neutral alumina of the crude bases (obtained by acid–base extraction) of a Kalir[2] synthetic mixture resulted in several fractions (in addition to the PCP-containing fraction), three of which proved to consist of a single component when examined by HPLC. The first of these, eluting before **2**, was identified as 1-[1-(phenylethyl)cyclohexyl]piperidine (**3**) (average yield 0.5%). The ^1H NMR spectrum (see Experimental Section) showed a quartet-doublet, indicating the CH_3CH unit. The mass spectrum exhibited a weak molecular ion at *m/e* 271 (2% base) and an M − 1^+ peak at 270 (5% base).[6] Fragments of *m/e* 166 (base peak) and 105 (50% base)

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