Table II—Pharmacokinetics of Lidocaine in a Patient following a Prolonged Infusion[a]

Hours	First Infusion, μg/ml	Second Infusion, μg/ml
−29.45	0	0
−17.45	2.98	2.85
0	4.04	4.55
1	3.39	3.15
2	2.28	2.39
4	1.43	1.72
8	0.35	0.54
12	0.20	0.31
19	0.10	—
20	—	0.10
23	N.D.[b]	N.D.

[a] The first and second infusions were separated by 2 weeks. [b] N.D. = not detectable.

ducibility of this analytical method and the difference in the pharmacokinetic parameters of lidocaine following a prolonged infusion. The two phases, α and β, and their values were virtually identical for the two slow infusions. The first infusion resulted in a $T_{p1/2}(\alpha)$ of 2.08 hr and a $T_{p1/2}(\beta)$ of 6.18 hr; the second infusion resulted in a $T_{p1/2}(\alpha)$ of 2.14 hr and a $T_{p1/2}(\beta)$ of 4.93 hr.

Therefore, it can be concluded that the analytical method described confirms that the pharmacokinetic parameters of lidocaine are identical for any given subject on the same treatment. However, compared to the healthy volunteers, the pharmacokinetic parameters were quite different. A true equilibrium was not reached, since the drug concentrations increased continuously during the infusion. This phenomenon can be explained by the fact that lidocaine inhibits its own metabolism *via* its acetylated metabolite (5); moreover, its renal excretion is slower, due to a reduction in the cardiac inotropic form and rhythm which significantly reduce blood flow. The marked lengthening of the α- and β-phases indicates that considerable caution should be exercised when lidocaine is administered by slow infusion, because the toxic effects with lidocaine are greater when the blood levels are too high.

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Kinetics and Mechanism of Blue Tetrazolium Reaction with Corticosteroids

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Abstract □ The kinetics of the reaction of blue tetrazolium with corticosteroids were investigated under pseudo-first-order conditions. The reaction rates for various corticosteroids were determined at various temperatures, and the enthalpy and entropy of activation for these compounds were determined. A mechanism is proposed in which an electron pair and a proton are transferred to blue tetrazolium from the anion formed by the action of tetramethylammonium hydroxide on the α-carbonyl moiety of the corticosteroid. The proposed mechanism is consistent with previous experimental results.

Keyphrases □ Blue tetrazolium—kinetics of reaction with various corticosteroids, effect of temperature, mechanism proposed □ Corticosteroids, various—kinetics of reaction with blue tetrazolium, effect of temperature, mechanism proposed □ Glucocorticoids, various—kinetics of reaction with blue tetrazolium, effect of temperature, mechanism proposed

Various forms of the blue tetrazolium reaction have been used for the quantitative determination of corticosteroids (1–4). The most widely used method is a slightly modified procedure of Mader and Buck (2), which is the official USP (5) and NF (6) method.

In strongly alkaline solution, blue tetrazolium [3,3′-[3,3′ - dimethoxy(1,1′-biphenyl)-4,4′-diyl]bis(2,5-diphenyl-2*H*-tetrazolium) dichloride] (I) oxidizes the α-carbonyl moiety of the C-17 side chain of the corticosteroid and is reduced quantitatively to a highly colored formazan whose concentration is measured spectrophotometrically. Extensive investigations (1, 3, 4, 7–16) of the reaction conditions established that the analytical procedure is subject to many variables, which are minimized by concurrently analyzing blank, standard, and sample.

Certain kinetic aspects of the blue tetrazolium reaction were reported, *e.g.*, the corticosteroid reaction is first order (17). Significant variations in the reaction rates of corticosteroids of closely related structures also were noted (1, 9, 14, 15, 18, 19). The reaction rate of corticosteroids with I was inversely related to the dielectric constant of the

solvating media in another study (20), and it was established that the α-carbonyl moiety of the corticosteroid reduces I (18).

Recently, a bimolecular scheme was proposed in which a "reduction unit" is transferred from the α-carbonyl moiety to I to produce a formazan (19); however, the reduction unit and the mode of transfer could not be ascertained. A free radical mechanism was proposed for the blue tetrazolium reaction (21), but this route has been disproven (19).

This paper reports the rate constants for selected corticosteroids at various temperatures and the obtained enthalpy and entropy of activation parameters. A mechanism consistent with the observed experimental data is proposed.

EXPERIMENTAL

Apparatus—A UV–visible recording spectrophotometer equipped with water-jacketed cell holders and 1-cm quartz cells[1], a microbalance[2], and a temperature-controlled circulating bath[3] were used.

Reagents—Alcohol USP and analytical grade absolute methanol were used as received. A 1% solution of tetramethylammonium hydroxide (II) was prepared. A 5.00-ml aliquot of II, 10% aqueous[4], was diluted to 50.0 ml with alcohol USP. Blue tetrazolium[5] (I), 5.0 mg/ml, was prepared in absolute methanol.

All corticosteroid standard solutions contained 0.010 mg/ml in alcohol USP.

General Procedure—The procedure, unless otherwise specified, is the official procedure given in the USP XIX (5), in which a 20.00-ml aliquot of standard or sample corticosteroid in alcohol USP is treated with 2.00 ml of I, 5.0 mg/ml, followed by 2.00 ml of 1% II. The absorbance is measured spectrophotometrically against a reagent blank 90 min after the II addition.

Rate Studies—A 20.00-ml aliquot of a standard corticosteroid and a 20.00-ml blank of alcohol USP were placed in jacketed cell holders and allowed to temperature equilibrate for 15 min. The aliquots were treated by the general procedure, with zero time taken as the time of addition of 1% II to the standard solution. Both solutions were transferred to cells as rapidly as possible and placed in the spectrophotometer, and absorbance readings were made each minute at 525 nm until the reaction reached essential completion. The procedure was repeated for five corticosteroids at five different temperatures.

RESULTS AND DISCUSSION

The results of the kinetics studies of the reaction of blue tetrazolium (I) with corticosteroids are shown in Table I. Pseudo-first-order rate constants were observed for all corticosteroids. The rate constants were obtained by performing a least-squares regression on the equation:

$$\ln (1 - A_i/A') = -k_r t_i + E \qquad \text{(Eq. 1)}$$

where A_i is the absorbance at time t_i, A' is the optimum absorbance, k_r is the observed rate constant, and E is the intercept. The correlation coefficient for all curves was greater than 0.99.

Figure 1 is a typical kinetic plot for prednisone. The value of A' was calculated from the average value of absorbance per micromole of formazan, 1.031 (19), and the initial micromolar concentration of the corticosteroid in the 20.00-ml aliquot. The estimation of A' was necessitated for two reasons. First, under the described reaction conditions, a mixture of two formazans of different geometrics and molar absorptivities is formed. The nature and properties of these formazans were investigated previously (20). Second, as the reaction time increases, base decomposition of the formazan begins, causing a decrease in the absorption maxima for the reaction.

To check the reliability of the results, the rate constants of certain corticosteroids were determined by the method developed by Guggenheim (22). The Guggenheim method was not applicable to all corticosteroid reactions because of the rapid completion and concurrent deviation from linearity of most of them at elevated temperatures. Some rates calculated from the Guggenheim method were within experimental accuracy of the previously obtained values (Table I).

Table I—Kinetic and Thermodynamic Parameters for the Reaction of Corticosteroids with Blue Tetrazolium

Corticosteroid	T, °K	k_r, min^{-1}
Dihydrocortisone[a]	303.77	2.809×10^{-1}
	303.76	2.708×10^{-1}
	303.39	3.116×10^{-1}
	300.38	2.030×10^{-1}
	296.41	1.333×10^{-1}
	292.48	1.103×10^{-1}
	287.49	5.775×10^{-2}
	285.80	4.552×10^{-2}
	284.13	3.450×10^{-2}
	283.95	3.608×10^{-2}
	283.87	3.421×10^{-2}
		1.146×10^{-1} [b]
		$\Delta H^{\ddagger} = 17.3$ kcal/mole
		$\Delta S^{\ddagger} = -12.2$ eu
Dexamethasone[c]	303.78	1.120×10^{-1}
	303.77	1.272×10^{-1}
	300.38	6.991×10^{-2}
	296.41	5.447×10^{-2}
	292.48	3.982×10^{-2}
	287.49	1.788×10^{-2}
	285.80	1.085×10^{-2}
	284.22	1.171×10^{-2}
	284.03	1.265×10^{-2}
		1.718×10^{-2}
		$\Delta H^{\ddagger} = 19.8$ kcal/mole
		$\Delta S^{\ddagger} = -5.7$ eu
Prednisone[d]	303.55	3.876×10^{-1}
	303.46	3.202×10^{-1}
	300.38	2.322×10^{-1}
	296.41	1.959×10^{-1}
	292.48	1.373×10^{-1}
	287.49	7.128×10^{-2}
	285.80	4.277×10^{-2}
	284.24	4.698×10^{-2}
	283.87	4.690×10^{-2}
	283.82	4.699×10^{-2}
		$\Delta H^{\ddagger} = 17.3$ kcal/mole
		$\Delta S^{\ddagger} = -11.7$ eu
Corticosterone[a]	303.72	1.320×10^{-1}
	303.71	1.211×10^{-1}
	300.38	7.025×10^{-2}
	296.41	6.324×10^{-2}
	292.48	3.880×10^{-2}
	287.49	2.115×10^{-2}
	285.80	1.096×10^{-2}
	283.74	1.286×10^{-2}
	283.70	1.236×10^{-2}
	283.68	1.314×10^{-2}
		2.207×10^{-2} [b]
		$\Delta H^{\ddagger} = 19.1$ kcal/mole
		$\Delta S^{\ddagger} = -8.1$ eu
5-Pregnene-3β,21-diol-20-one[a]	303.74	8.709×10^{-2}
	303.71	8.853×10^{-2}
	300.38	5.532×10^{-2}
	296.41	4.638×10^{-2}
	292.48	3.260×10^{-2}
	287.49	1.889×10^{-2}
	285.80	1.182×10^{-2}
	283.78	1.075×10^{-2}
	283.70	1.046×10^{-2}
	283.59	1.087×10^{-2}
		2.888×10^{-2} [b]

[a] Rates determined by method of Guggenheim (22). [b] Schwarz/Mann, Orangeburg, N.Y. [c] NF reference standard. [d] USP reference standard.

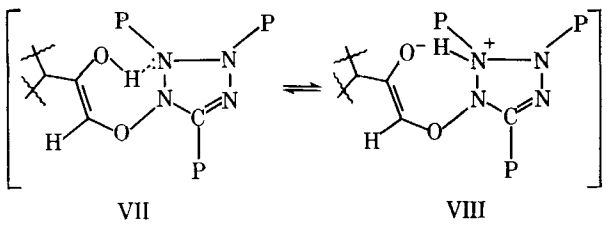
The enthalpy of activation and entropy of activation results are also shown in Table I. The values were obtained by performing a least-squares regression on the equation:

$$\ln (k_r h/kT) = -\Delta H^{\ddagger}/RT + \Delta S^{\ddagger}/R \qquad \text{(Eq. 2)}$$

where k_r is the observed rate constant, h is Planck's constant, k is Boltzman's constant, T is absolute temperature, R is the gas constant, and $\Delta H^{\ddagger}$ and $\Delta S^{\ddagger}$ are the enthalpy and entropy of activation, respectively. The correlation coefficient was greater than 0.95 in all cases.

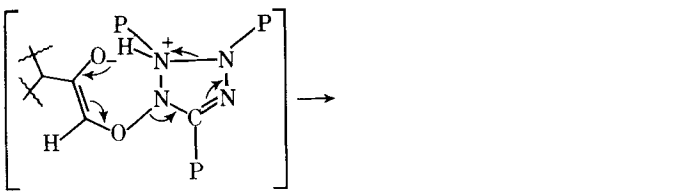
[1] Cary model 118.
[2] Mettler model M.
[3] Neslab model RTE-3.
[4] Eastman Chemical Co.
[5] Dajae Laboratories.

IV + VI ⇌

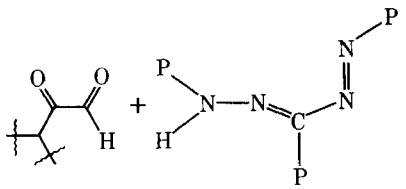


VII VIII

Scheme I



VIII

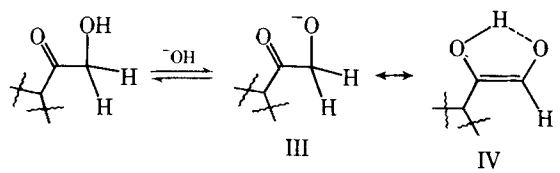


Scheme II

From these data and from previous observations (18, 19), the reactivity of the corticosteroids toward I appears to be, in part, a function of their molecular shape. Steroids with C-16 substitution in either the α- or β-position react the slowest, steroids that possess a planar A ring and have sp^3 C-11 substitution show intermediate reactivity, and steroids possessing a C-11 carbonyl react the fastest with I. These observations strongly imply that the approach of the I molecules occurs at the β-face of the steroid since the greatest rate retardation is observed for steroids having β-substitution near the carbonyl group.

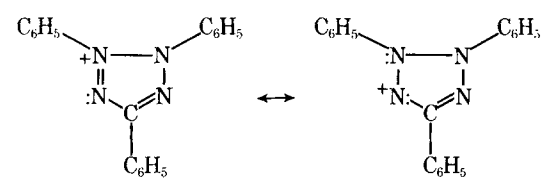
The mechanism proposed for the reduction of I by corticosteroids is shown in Schemes I and II and involves the formation of a complex (VII ⇌ VIII) between the enolate ion, IV, of the corticosteroid molecule and I, followed by a transfer of an electron pair and a proton from the enolate portion of the complex to a positively charged nitrogen atom on the I portion to produce a glyoxal derivative and a formazan (IX). The glyoxal then decomposes to the various products characterized by Lewbart and Mattox (23, 24).

Compounds containing α-carbonyl groups undergo rearrangement in basic solution *via* an enediol intermediate (25–27); in some cases, the enediol itself is the more stable form (28). With regard to the enolate ion, a species such as IV would appear to be more stable than III because of its ability to exist in a cyclic five-membered species having intramolecular hydrogen bonding (Scheme III). In a basic solution, II exists as a resonance hybrid, in which V and VI (Scheme IV) are important contributing structures. The species VI would seem to contribute more to the hybrid



III IV

Scheme III



V VI

Scheme IV

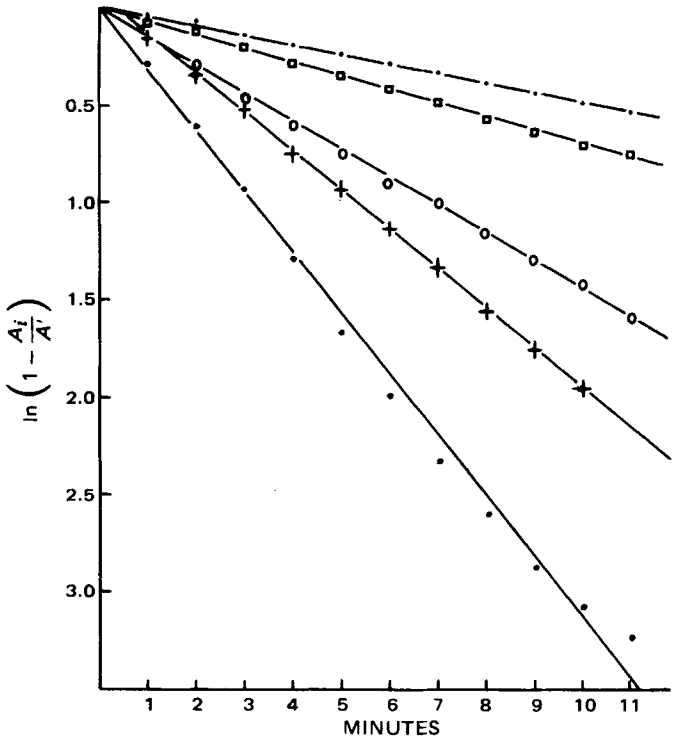


Figure 1—*Reaction rate of a typical corticosteroid at various temperatures. Key: ·, 10°; □, 15°; ○, 20°; +, 25°; and ●, 30°.*

than V since the lone pair of electrons of the nitrogen in VI can be further delocalized into the phenyl group.

The mechanism is in accordance with the observed experimental data. Bimolecular reactions involving a well-oriented cyclic transition complex generally have an entropy of activation value of from −5 to −15 eu simply as a result of the entropy requirement for bringing together two molecules to form a single complex (29). Thus, observed values of $\Delta S^{\ddagger}$ for the reaction of I with corticosteroids provide further evidence of the bimolecularity of the reaction path. The enthalpy of activation values are also consistent with results generally observed for bimolecular reactions in solution (30).

Furthermore, this mechanism predicts that an increase in the dielectric constant would decrease the intermediate complex because of the ionic nature of the reaction and, hence, decrease the reaction rate. This conclusion is in accordance with the observations of Graham *et al.* (20).

Construction of a Dreiding model of the intermediate complex exemplifies and supports the β-face attack and its subsequent decrease of rate. Also, if one follows the complex breakdown with the model, the geometry of the species formed is seen to be identical with that reported as the major formazan isomer (20).

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Dosage Form Index: An Objective Criterion for Evaluation of Controlled-Release Drug Delivery Systems

F. THEEUWES ˣ and W. BAYNE

Abstract □ A dimensionless parameter, the dosage form index (DI_τ), is proposed for evaluating the performance of drug delivery systems. The index is defined as the ratio of the maximum to minimum concentrations of the drug in plasma within each interdose interval (in hours), τ, during repetitive administration of the dosage form in the quasisteady state. Dosage form indexes can be averaged among subjects or within subjects at successive time periods to arrive at a mean value. As an example, two GI therapeutic systems—the 15- and 20-mg/hr acetazolamide systems that deliver drug at constant rates for 6 and 12 hr and contain 125 and 250 mg, respectively—were compared in normal subjects with a commercial sustained-release product containing 500 mg of acetazolamide. The dosage form index, DI_{24}, was 4.9 for the sustained-release dosage form and 3.2 for the 20-mg/hr system; DI_{12} was 1.6 for the 15-mg/hr system.

Keyphrases □ Drug delivery systems—sustained-release product and 15- and 20-mg/hr GI acetazolamide systems compared in humans, dosage form index suggested as evaluating parameter □ Dosage form index—suggested as evaluating parameter for drug delivery systems, sustained-release product and 15- and 20-mg/hr GI acetazolamide systems compared in humans □ Acetazolamide—sustained-release product and 15- and 20-mg/hr GI drug delivery systems compared in humans, dosage form index suggested as evaluating parameter □ Carbonic anhydrase inhibitors—acetazolamide, sustained-release product and 15- and 20-mg/hr GI drug delivery systems compared, humans

Optimum therapy with repetitive administration of conventional dosage forms (*e.g.*, injectables, liquids, or tablets) can classically be pursued by dosage scheduling. The aim of this process is to maintain drug concentrations in a therapeutic range, above the minimum effective concentration and below the toxic concentration.

The time course of systemic drug concentrations following repetitive dosing of intravenous injections and tablets, based on different absorption and elimination rate constants, was described mathematically using one-compartment and multicompartment pharmacokinetic models (1–3).

BACKGROUND

To discuss optimum drug dosing, the therapeutic index (TI) should be defined in terms of plasma concentrations (4) rather than dose (5):

$$\mathrm{TI} = \frac{C^*_{max}}{C^*_{min}} \qquad \text{(Eq. 1)}$$

where C^*_{max} and C^*_{min} are the maximum and minimum desired plasma concentrations, respectively, and must be defined further for each specific drug in relation to pharmacological responses.

Dosage scheduling should be carried out at a frequency such that the ratio of maximum to minimum plasma concentrations is less than TI and at a dose of sufficient magnitude to yield effective levels.

For a linear, one-compartment system with repetitive intravenous injections at pseudo-steady state, the ratio of maximum to minimum plasma concentrations is given by:

$$\frac{C_{max}}{C_{min}} = e^{k\tau} \qquad \text{(Eq. 2)}$$

where k is the drug elimination rate constant, and τ is the dosing interval. The C_{max}/C_{min} ratio has been called (1) Q_{ext}, the ratio of asymptotic extreme concentrations within a dosing interval of the multiple-dose curve. When the therapeutic index (TI) is known, the proper dosage interval must then be dictated by:

$$e^{k\tau} \leq \mathrm{TI} \qquad \text{(Eq. 3)}$$

or:

$$\tau \leq t_{1/2}\frac{\ln \mathrm{TI}}{\ln 2} \qquad \text{(Eq. 4)}$$

For TI = 2, the dosing interval should maximally be equal to the biological half-life of the drug. For drugs with a half-life of less than 12 hr and a low therapeutic index, proper dosage schedules are inconveniently frequent. To alleviate these problems, therapeutic systems were developed (6–8).

Therapeutic systems are a class of dosage forms designed to improve drug therapy through controlled administration of drug substances, in time and space, to elicit the optimum sum total of pharmacological response. They are specified not by content, size, or shape but by function: the rate at which they deliver drug *in vivo* and the duration for which they do so.