Chemical Equilibria in Solutions of Bisulfite Salts

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Abstract □ The equilibrium between bisulfite ion and pyrosulfite ion in aqueous solutions of different ionic strengths was investigated spectrophotometrically. Values for the equilibrium constant at ionic strengths of 2.0, 0.9, 0.5, 0.1, and 0.01 *M* were found to be 0.34, 0.22, 0.175, 0.115, and 0.086 M^{-1}, respectively. The extrapolated thermodynamic equilibrium constant was calculated to be 0.076 ± 0.010 M^{-1}, and the molar absorptivity of pyrosulfite ion at 255 nm was calculated to be 1980. These values differ significantly from the values in the chemical literature. No evidence was found for the formation of complexes between bisulfite and citrate ions in aqueous solution.

Keyphrases □ Bisulfite ion-pyrosulfite ion—equilibrium constants, complex formation with citrate ions □ Equilibrium constants—bisulfite-ion-pyrosulfite-ion equilibrium □ Complex formation—bisulfite ion, citrate □ Citrate ions—complex formation with bisulfite ions

When sulfur dioxide or salts of sulfurous acid (hydrated SO_2, H_2SO_3) are dissolved in water, the following equilibria are rapidly established[1]:

$$H_2SO_3 \overset{K_a^1}{\rightleftharpoons} HSO_3^- + H^+$$

$$HSO_3^- \overset{K_a^2}{\rightleftharpoons} SO_3^{-2} + H^+$$

$$2HSO_3^- \overset{K}{\rightleftharpoons} S_2O_5^{-2} + H_2O$$

Scheme I

Therefore, accurate values of the equilibrium constants K_a^1, K_a^2, and K must be known to calculate the relative concentrations of sulfurous acid, bisulfite ion (HSO_3^-), sulfite ion (SO_3^{-2}), and pyrosulfite ion ($S_2O_5^{-2}$) in any given solution. Even though bisulfite salts have been used for many years in the pharmaceutical, food, and beverage industries, and the chemical reactivity of bisulfite-sulfite salt solutions has been widely studied[2], the current literature (1, 3, 4) contains conflicting values for the equilibrium constant K (Table I).

One aim of the present study was to determine values of concentration equilibrium constants for the dimerization of bisulfite ion at various ionic strengths and to use this information to calculate a thermodynamic equilibrium constant for this reaction.

In addition, an investigation was undertaken to see whether citrate ions reduce the thermodynamic activity of bisulfite ion when the two are included in aqueous solutions. It was reported (5) that citrate and bisulfite ions may form a complex in solution. If this was true, the reactivity of bisulfite ion in such solutions would be changed, and this may have serious consequences on its effectiveness as an antioxidant or as a participant in other reactions.

Table I—Equilibrium Constants for the Reaction:

$$2HSO_3^- \overset{K}{\rightleftharpoons} S_2O_5^{-2} + H_2O$$

and Spectral Characteristics of Pyrosulfite Ion

K, M^{-1}	ϵ^a	λ_{max}	T	$K\epsilon$	Ionic Strength, M	Reference
0.07	4000	255	20°	288	—[b]	1
2.0	142.9	256–258	22°	286	0.3	3
0.07[c]	2527	255	25°	177	—[b]	4
0.34	1980	255	25°	673	2.0	This work
0.076	1980	255	25°	139	0.0	This work

[a] Molar absorptivity of pyrosulfite ion at λ_{max}. [b] Ionic strength not indicated. [c] From Ref. 1.

EXPERIMENTAL

Materials and Apparatus—Sodium bisulfite, acetic acid, sodium chloride, hydrochloric acid, sodium hydroxide, citric acid, and sodium sulfite were all reagent grade. 1,2-Dihydro-2-imino-1-methylpyrimidine hydrochloride was prepared as described previously (6), mp 277–278°. All solutions were made with double-distilled water through which oxygen-free nitrogen had been bubbled for 1 hr. One percent of methanol was added to stabilize sodium bisulfite against oxidation. Final bisulfite concentrations were determined by iodometric titration.

Spectrophotometric measurements were made on three spectrophotometers[3]. Quartz cells of path length 1, 2, 10, 20, 50, and 100 mm were used for UV measurements. Temperature was maintained for UV readings, determinations of ionization constants, and kinetic runs at 25.0 ± 0.1°. The pH values were measured using a pH meter[4] standardized with U.S. National Bureau of Standards quality buffers at pH 4.00 and 7.00.

Ionization Constants—The two pKa values of sulfurous acid were measured by potentiometric titration using a method described previously (7). For pK_a^1, a 0.1 *M* solution of sulfurous acid was titrated with 1.0 *M* NaOH solution. The second ionization constant was obtained by titrating a 0.01 *M* solution of sodium bisulfite with 0.2 *M* NaOH. The ionic strength was maintained at 2.0 *M* with sodium chloride.

Molar Absorptivity—The molar absorptivity of H_2SO_3 was determined in 1 *M* HCl with 1 *M* NaCl added to maintain ionic strength at 2.0 *M*. The molar absorptivity of SO_3^{-2} was determined using solutions of sodium sulfite of 2.0 *M* total ionic strength.

Kinetic Method—The reaction of 1,2-dihydro-2-imino-1-methylpyrimidine hydrochloride with bisulfite ion (0.005 *M*) was followed at 302 nm for 2–3 half-lives and found to follow first-order kinetics. The pH was maintained at 4.14 ± 0.05 with 0.2 *M* acetate buffer or with 0.1, 0.2, or 0.3 *M* citrate buffer. Ionic strength was maintained at 0.5 *M* with sodium chloride.

Iodometric Titration—Five milliliters of a sodium bisulfite solution (0.055 *M*) was added to 10 ml of 0.1 *N* iodine solution and

[1] The presence of a further species, $HO{\cdot}SO_2^-$, was suggested (1) and refuted (2); since the extent of the reaction $HSO_3^- \rightleftarrows HO{\cdot}SO_2^-$ is independent of pH and concentration and neither species absorbs light strongly at the wavelengths of interest in the present study (above 230 nm), this species is not discussed in this report.

[2] The chemistry of sulfur dioxide was comprehensively reviewed by L. C. Schroeter in "Sulfur Dioxide," Pergamon, New York, N.Y., 1966.

[3] Cary 14, 15, and 16.

[4] Radiometer model 26.

Table II—Molar Absorptivity of Sulfur Species in Aqueous Sodium Bisulfite Solutions

Species	ϵ_{255}
H_2SO_3	450
HSO_3^-	<1
SO_3^{-2}	9.36
$S_2O_5^{-2}$	1980

titrated with 0.1 *N* sodium thiosulfate. In some experiments, 1, 5, or 15 ml of a 1 *M* sodium citrate solution, pH ≃ 4.0, was added to the sodium bisulfite solution before the addition of the iodine solution.

RESULTS AND DISCUSSION

Bisulfite-Pyrosulfite Equilibrium—The species present in aqueous solutions of sodium bisulfite (HSO_3^-, H_2SO_3, and SO_3^{-2}) all absorb UV light but different amounts at different wavelengths. This difference at 255 nm (Table II) permits one to investigate the composition of aqueous solutions of sodium bisulfite spectrophotometrically. Since bisulfite ion itself absorbs very little light at 255 nm, the absorbance at this wavelength is due almost entirely to pyrosulfite ion, sulfurous acid, and sulfite ion. Hence:

$$A_T \simeq \epsilon_{255}(S_2O_5^{-2})[S_2O_5^{-2}]_e + \epsilon_{255}(H_2SO_3)[H_2SO_3]_e + \epsilon_{255}(SO_3^{-2})[SO_3^{-2}]_e \quad \text{(Eq. 1)}$$

where A_T is the total absorbance at 255 nm; ϵ_{255} is the molar absorptivity at 255 nm of the species indicated; and $[S_2O_5^{-2}]_e$, $[H_2SO_3]_e$, and $[SO_3^{-2}]_e$ are the equilibrium concentrations of $S_2O_5^{-2}$, H_2SO_3, and SO_3^{-2}, respectively. If S_T is the total sulfur concentration as determined iodometrically, then:

$$S_T = [H_2SO_3]_e + [SO_3^-]_e + [SO_3^{-2}]_e + 2[S_2O_5^{-2}]_e \quad \text{(Eq. 2)}$$

Using the equations defining the equilibria in Scheme I:

$$K_a^1 = [H^+][HSO_3^-]/[H_2SO_3] \quad \text{(Eq. 3)}$$

$$K_a^2 = [H^+][SO_3^{-2}]/[HSO_3^-] \quad \text{(Eq. 4)}$$

$$K = [S_2O_5^{-2}]/[HSO_3^-]^2 \quad \text{(Eq. 5)}$$

and substituting in Eq. 2 give:

$$S_T = [HSO_3^-]_e \frac{[H^+]^2 + K_a^1[H^+] + K_a^1K_a^2}{K_a^1[H^+]} + 2K[HSO_3^-]_e^2 \quad \text{(Eq. 6)}$$

This equation can then be solved for $[HSO_3^-]_e$ if a value of K is assumed. Values of $[S_2O_5^{-2}]_e$, $[SO_3^{-2}]_e$, and $[H_2SO_3]_e$ can be calculated from Eqs. 3–5, and a value of A_T can be calculated from Eq. 1 by assuming a value of ϵ_{255} ($S_2O_5^{-2}$). The experimental results in Table III were obtained using a wide range of values of S_T

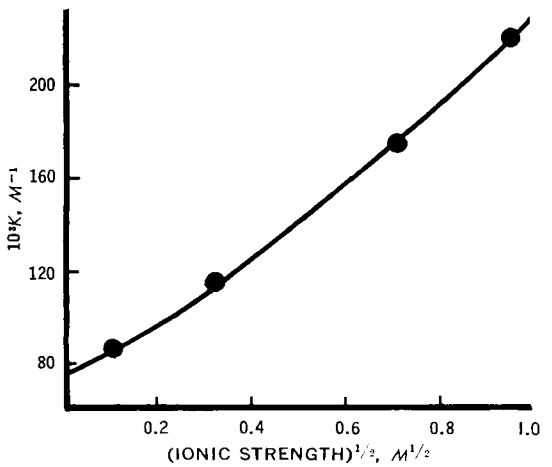

Figure 1—*Plot of values of* K *at 25° against (ionic strength)*$^{1/2}$.

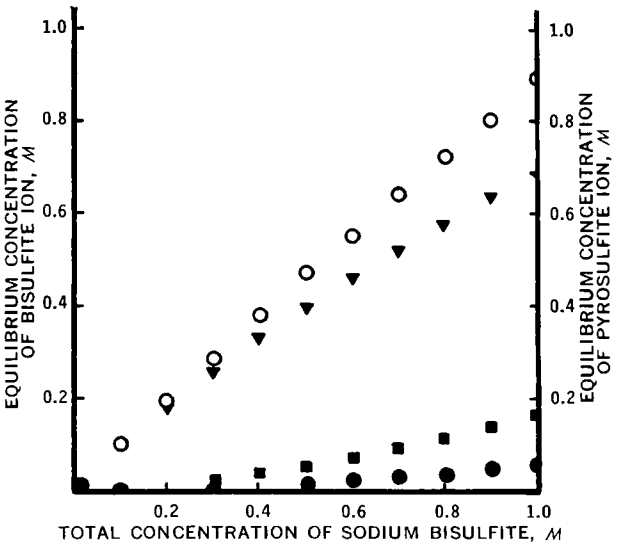

Figure 2—*Plots against total sodium bisulfite concentrations of the equilibrium concentrations of bisulfite ion and pyrosulfite ion that would be calculated if it were assumed that* K = *0.076* M^{-1} *and* K = *0.34* M^{-1}. *Key:* ○, $[HSO_3^-]_e$, *when* K = *0.076* M^{-1}; ▼, $[HSO_3^-]_e$, *when* K = *0.34* M^{-1}; ●, $[S_2O_5^{-2}]_e$, *when* K = *0.076* M^{-1}; *and* ■, $[S_2O_5^{-2}]_e$, *when* K = *0.34* M^{-1}.

(0.005–0.16 *M*) and pH values from 3.0 to 6.0. Total ionic strength was maintained constant at 2.0 *M* with sodium chloride.

The values of K_a^1, K_a^2, ϵ_{255} (SO_3^{-2}), and ϵ_{255} (H_2SO_3) were obtained independently, and values of K and ϵ_{255} ($S_2O_5^{-2}$) were changed by trial and error to obtain a minimum value for the sum of the square of the deviations between calculated absorbance and measured absorbance. At an ionic strength of 2.0 *M*, values of K = 0.34 M^{-1} and ϵ_{255} ($S_2O_5^{-2}$) = 1980 gave the best fit.

In dilute solutions of sodium bisulfite at pH values where there is little sulfurous acid or sulfite ion, about pH 4.0, the concentration of bisulfite ion is approximately equal to S_T and Eq. 1 simplifies to:

$$A_T = K\epsilon_{255}(S_2O_5^{-2})S_T^2 \quad \text{(Eq. 7)}$$

Values of $K\epsilon_{255}$ ($S_2O_5^{-2}$) can thus be obtained by plotting A_T *versus* S_T^2. In this way, values of $K\epsilon_{255}$ ($S_2O_5^{-2}$) at pH 4.00 ± 0.01 and ionic strengths (μ) from 0.01 to 0.9 *M* were calculated. Because the value of ϵ_{255} ($S_2O_5^{-2}$) is independent of ionic strength, values of K at these ionic strengths could then be calculated. Results are shown in Fig. 1 as a plot of K against $\sqrt{\mu}$. From this figure, the value of K at zero ionic strength was obtained by extrapolation. This value was 0.076 ± 0.010 M^{-1}.

Previously calculated values for K are shown in Table I. The values of Golding (1) and Eriksen and Lind (4) suffer from the fact that ionic strength was apparently not maintained constant.

Golding (1) assumed $[H_2SO_3]_e$ and $[SO_3^{-2}]_e$ to be approximately zero under his experimental conditions and substituted Eq. 5 into Eq. 2 to derive Eq. 8:

$$S_T/A_T = X[A_T^{-1/2}] + Y \quad \text{(Eq. 8)}$$

where $X^2 = [\epsilon_{255}\ (S_2O_5^{-2})\ (d)\ (K)]^{-1}$, and $Y = 2/[\epsilon_{255}\ (S_2O_5^{-2})\ (d)]$.

In these identities, d (in centimeters) was the path length of the spectrophotometer cell. Hence, plots of S_T/A_T against $A_T^{-1/2}$ were used to calculate a value of $K\epsilon_{255}$ ($S_2O_5^{-2}$) and ϵ_{255} ($S_2O_5^{-2}$). Because the value of the intercept was very small, the error in the value of ϵ_{255} ($S_2O_5^{-2}$) was large. Since K was obtained from the term $K\epsilon_{255}$ ($S_2O_5^{-2}$), an error of equal magnitude was thus carried into the value determined for K. The value of K of Arkhipova and Chistyakova (3) at an ionic strength of 0.3 was computed by trial-and-error minimizing of deviations between experimental and calculated values of absorbance. However, these workers used quite a narrow range (0.045–0.06 *M*) of S_T values, so considerable error could be involved in the value of K they obtained.

Table III—Absorbance [Experimental[a] (A_{exp}) and Calculated[b] (A_{calc})] of Aqueous Sodium Bisulfite $[Bis]_T$ Solutions with Ionic Strength 2.0 M Adjusted with Sodium Chloride

$[Bis]_T$ = 0.160 M				Cell Length 1 mm				
pH	5.90	5.64	5.34	4.92	4.48	4.05	3.60	3.16
A_{exp}	0.735	0.910	1.108	1.260	1.355	1.455	1.520	1.602
A_{calc}	0.694	0.919	1.124	1.292	1.370	1.404	1.430	1.473
$[Bis]_T$ = 0.079 M				Cell Length 2 mm				
pH	5.95	5.68	5.36	4.94	4.50	4.04	3.53	3.12
A_{exp}	0.340	0.450	0.515	0.630	0.665	0.720	0.780	0.850
A_{calc}	0.356	0.474	0.590	0.682	0.728	0.752	0.782	0.836
$[Bis]_T$ = 0.400 M				Cell Length 10 mm				
pH	5.86	5.65	5.34	4.93	4.41	3.92	3.42	
A_{exp}	0.560	0.635	0.758	0.855	0.960	1.020	1.125	
A_{calc}	0.567	0.670	0.799	0.907	0.973	1.017	1.110	
$[Bis]_T$ = 0.019 M				Cell Length 20 mm				
pH	5.81	5.72	5.42	4.99	4.51	4.01	3.50	3.03
A_{exp}	0.310	0.328	0.358	0.412	0.448	0.493	0.598	0.834
A_{calc}	0.330	0.342	0.384	0.428	0.458	0.488	0.564	0.768
$[Bis]_T$ = 0.011 M				Cell Length 50 mm				
pH	5.69	5.56	5.39	5.00	4.52	4.00	3.51	3.05
A_{exp}	0.308	0.308	0.310	0.334	0.371	0.410	0.521	0.852
A_{calc}	0.327	0.312	0.337	0.353	0.371	0.408	0.504	0.785
$[Bis]_T$ = 0.005 M				Cell Length 100 mm				
pH	5.66	5.50	5.30	4.97	4.46	3.94	3.47	3.00
A_{exp}	0.208	0.190	0.162	0.157	0.137	0.196	0.320	0.672
A_{calc}	0.204	0.191	0.181	0.173	0.177	0.212	0.315	0.612

[a] Absorbance at 255 nm. [b] Absorbance calculated using K = 0.34 M^{-1} and ϵ_{255} ($S_2O_5^{-2}$) = 1980.

The importance of having a reliable value of K when estimating the effective concentration of bisulfite ion or pyrosulfite ion in a solution of sodium bisulfite is illustrated in Fig. 2. This figure displays plots of $[HSO_3^-]_e$ and $[S_2O_5^{-2}]_e$ against $[HSO_3^-]_T$ that would be calculated if K values of 0.076 M^{-1} (the actual value at zero ionic strength) or 0.34 M^{-1} (the actual value at an ionic strength of 2.9 M) were used in the calculation.

The ionic strength dependence of K values indicates that, although dilute solutions of sodium bisulfite contain very little pyrosulfite ion if the ionic strength is low, the concentration of pyrosulfite ion can become appreciable if the ionic strength is increased.

Bisulfite-Citrate-Ion Interaction—Formation of a bisulfite-citrate-ion complex has been suggested as a reason for the greater effectiveness of citrate buffers, at pH values near 4, in removing sulfur dioxide from waste gases (5) compared to other inorganic and mixed organic and inorganic solutions. However, no evidence to support this suggestion was found in the present study. As shown earlier, the UV spectrum of pyrosulfite ion is a sensitive measure of the bisulfite-ion activity in solution, and its intensity should change as the concentration of citrate is increased if a bisulfite-citrate complex is forming in solution. However, the absorbance at 255 nm of a sodium bisulfite solution (0.01 M) at constant pH (4.0) and ionic strength (1.5 M with sodium chloride) was unaffected by the addition of sodium citrate (0.05–0.5 M) when the contribution of the citrate ion itself was taken into account. The citrate ion apparently did not affect the activity of bisulfite ion as measured spectrophotometrically.

If a stable complex between citrate and bisulfite ion is formed and the rate of its dissociation is slow, then rapid iodometric titration of bisulfite-ion solutions containing citrate ion should give results different from those obtained in the absence of citrate ion. This was not found when a solution of sodium bisulfite (0.055 M) at pH 4.0 was titrated iodometrically in the presence of different amounts of citrate ion (0.05–1.0 M). Hence, a stable, slowly dissociating complex is not likely.

The reaction of bisulfite ion with 1,2-dihydro-2-imino-1-methylpyrimidine has been extensively studied (8), and the rate of reaction was found to be first order in bisulfite ion. If a complex is formed with citrate ion, the activity of bisulfite ion would be reduced, and its rate of reaction with 1,2-dihydro-2-amino-1-methylpyrimidine would be accordingly reduced.

This reaction was investigated in solutions of constant ionic strength (0.5 M), pH (4.0), and total bisulfite concentration (0.01 M) and different citrate-ion concentrations (0.1–0.3 M). The rate of reaction was found to be independent of citrate concentration. This kinetic result further indicates that a complex between citrate and bisulfite ion is unlikely.

The efficiency of citrate buffers in the removal of sulfur dioxide from waste gases could be due to citric acid being a good buffer, because it is polyprotic, having three ionizable hydrogens. As sulfur dioxide dissolves, it is rapidly hydrated to H_2SO_3, which can ionize to HSO_3^-; since HSO_3^- has a much higher solubility than H_2SO_3, the overall solubility of sulfur dioxide is increased. Since this ionization of H_2SO_3 is pH dependent, the increased buffer capacity of the citrate buffer will maintain a higher amount of the sulfur dioxide in the form of bisulfite ion, thereby increasing the overall solubility of sulfur dioxide. Also, because citric acid is polyprotic, its solutions at pH values around 4 would have relatively high ionic strengths compared to those of monoprotic acids. Consequently, the formation of pyrosulfite from bisulfite would be favored and the effective solubility of SO_2 would increase. Therefore, the efficiency of the citrate process can be explained without having a bisulfite-citrate-ion complex.

CONCLUSION

The literature values for the equilibrium constant for the dimerization reaction of bisulfite ion to pyrosulfite ion were conflicting. A study has been completed to clarify the situation by providing a reliable value of K, the ionic strength dependence of K, and the extinction coefficient of pyrosulfite ion at 255 nm. A thermodynamic equilibrium constant of 0.076 ± 0.010 M^{-1} at 25° and a molar absorptivity for $S_2O_5^{-2}$ at 255 nm of 1980 have been determined.

Studies on aqueous solutions of bisulfite ion and citrate ions have shown that a stable bisulfite-ion–citrate-ion complex is unlikely to be formed in appreciable amounts and that the thermodynamic activity of bisulfite ion is unaffected by the presence of citrate ions.

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Comparative Pharmacokinetics of Coumarin Anticoagulants X: Relationship between Distribution, Elimination, and Anticoagulant Action of Warfarin

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Abstract □ The biological half-life of warfarin in a group of male Sprague–Dawley rats varied from 5 to 28 hr, and the apparent volume of distribution varied from 102 to 320 ml/kg body weight. There was a strong and statistically highly significant positive correlation between the elimination rate constant and the apparent volume of distribution of warfarin in individual rats. There was no relationship between the elimination rate constant for warfarin and either the normal (prewarfarin) prothrombin complex activity or the rate constant for decline of this activity when the synthesis of vitamin K-dependent clotting factors was blocked. The concentration of warfarin in plasma at which the synthesis rate of prothrombin complex activity was inhibited by 50% varied from 0.05 to 1.37 μg/ml and showed a strong and highly statistically significant positive correlation with the biological half-life of warfarin in individual animals. These results demonstrate a pronounced association among the apparent volume of distribution, the elimination rate constant, and the plasma concentration–anticoagulant effect relationship for warfarin which may have to be considered in the design of clinical dosage regimens.

Keyphrases □ Anticoagulants, coumarin—comparative pharmacokinetics of warfarin, relationships between distribution, elimination, and activity, biological half-life, volume of distribution □ Coumarin anticoagulants—comparative pharmacokinetics of warfarin, relationships between distribution, elimination, and activity, biological half-life, volume of distribution □ Warfarin—comparative pharmacokinetics, relationships between distribution, elimination, and activity, biological half-life, volume of distribution □ Pharmacokinetics—warfarin distribution, elimination, and activity

Warfarin is probably the most widely used oral anticoagulant. Its biological half-life varies markedly between subjects. For example, the biological half-life of warfarin in 20 healthy adult human subjects was reported to range from 15 to 80 hr (1, 2), and there are case reports of patients with a warfarin half-life of only 5.5 and 6.1 hr (3, 4). In a study (5) of warfarin elimination by mongrel dogs, half-lives of 21–48 hr were found. In random-bred Sprague–Dawley rats, warfarin half-lives from <5 to 70 hr in males and from 10 to 90 hr in females have been observed (6).

It is generally believed that intersubject differences in pharmacological response to a particular dosage regimen of a drug due to differences in the rate of elimination of that drug may be overcome by adjusting the dosage regimen to attain a given plasma concentration, *i.e.*, the therapeutic concentration. This assumes, for example, that individuals who differ with respect to the half-life for warfarin elimination nevertheless respond equally (at least in the absence of certain other drugs and diseases) to a particular concentration of that drug in the plasma[1]. This assumption would be incorrect if the differences in the elimination kinetics of warfarin are caused by, or associated with, differences in the distribution of that drug in the body. In this study, the apparent volume of distribution, the biological half-life, and the plasma concentration–anticoagulant effect curve for warfarin in individual rats were determined and the relationships between these characteristics were examined.

EXPERIMENTAL

Male Sprague–Dawley rats[2], weighing 305–400 g, were used. They had unrestricted access to food[3] and water before and during the experiments. The animals received 0.6 mg ^{14}C-warfarin/kg body weight ip. Blood samples (0.36 ml) were obtained repeatedly from the tail artery (9) for up to 96 hr after drug administration. Each blood sample was mixed with 0.04 ml sodium oxalate

[1] There are rare instances of hereditary resistance to warfarin and other coumarin anticoagulants, but the distribution and elimination of warfarin in individuals so affected are entirely normal (7, 8).

[2] Blue Spruce Farms, Altamont, N.Y.

[3] Charles River formula 4RF.