KINETICS OF MUTAROTATION OF ALDOSES IN THE PRESENCE OF METALLIC IONS

By

Wesley Brock Neely

A THESIS

Submitted to the School of Graduate Studies of Michigan State College of Agriculture and Applied Science in partial fulfillment of the requirements for the degree of

DOCTOR OF PHILOSOPHY

Department of Chemistry

1952

KINETICS OF MUTAROTATION OF ALDOSES IN THE PRESENCE OF METALLIC IONS

By

Wesley Brock Neely

AN ABSTRACT

Submitted to the School of Graduate Studies of Michigan State College of Agriculture and Applied Science in partial fulfillment of the requirements for the degree of

DOCTOR OF PHILOSOPHY

Department of Chemistry

Year 1952

Approved John C. Speck, Jr.

Wesley Brock Neely

THESIS ABSTRACT

The mutarotation of aldoses in salt solutions was investigated and catalysis by lithium, beryllium, magnesium, calcium, cupric and ferric ions was observed. Catalytic constants were determined for each of these species.

Addition of the aforementioned ions to acetate buffers caused diminution of the observed rate constant at lower concentrations, whereas at higher concentration augmentation of the constant occurred. This phenomenon is explained by assuming that the acetates of these ions are incompletely dissociated. On this basis it is possible to treat the lithium acetate system mathematically. Two different mathematical analyses, A and B were applied. In A it was assumed that the reaction is bimolecular throughout. In B termolecular processes were allowed. Although neither the expression derived in A nor that in B permitted exact agreement with the observed rate constants, better values for these constants were obtained in the latter analysis.

From the observation that third order kinetics may be applied to the metal ion catalysis of mutarotation, a mechanism is presented in which a concerted attack by the metal ion and the nucleophilic reagent appear in the rate determining step.

ACKNOWLEDGMENT

The author wishes to express his sincere gratitude to Dr. J. C. Speck, Jr. for his invaluable assistance; and to the Graduate Alumni Association for their financial aid in the form of a Fellowship.

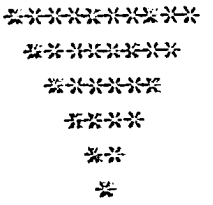


TABLE OF CONTENTS

HISTORICAL INTRODUCTION

HISTORICAL INTRODUCTION

The phenomenon of mutarotation of reducing sugars occupies an almost unique place in carbohydrate chemistry. Although it is of only minor importance in the synthesis of these substances and has no established role in any metabolic change or other biological event, its importance in the investigation of homogeneous catalysis renders it one of the most significant of all the transformations associated with carbohydrate substances.

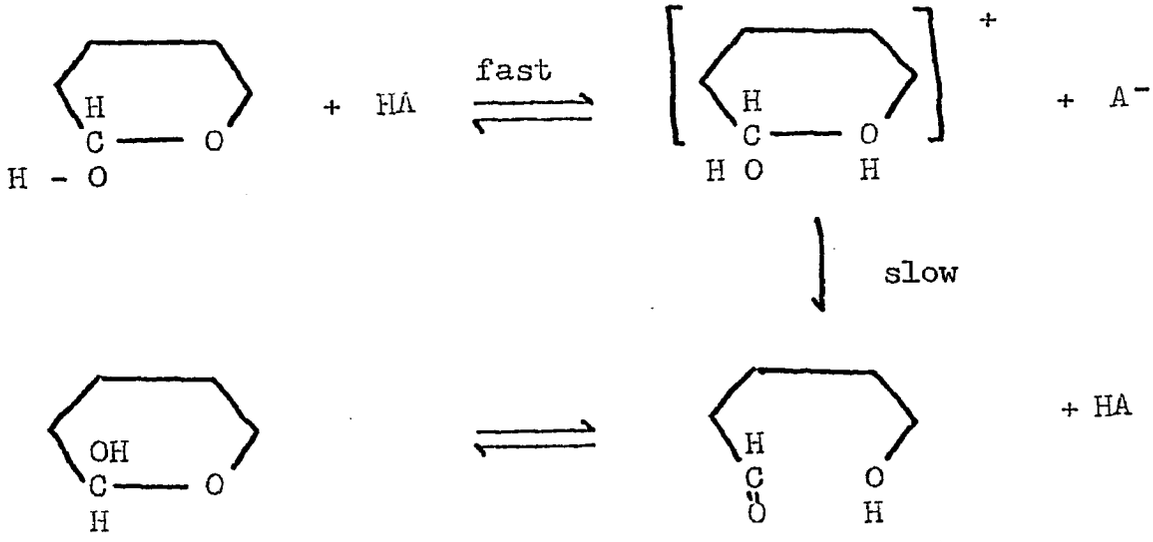
Mutarotation of a reducing sugar seldom appears to involve a simple change. Notwithstanding this, the α,β-inversion of the pyranose form is probably the correct mechanism for the mutarotation of glucose as ordinarily observed, as well as that for many of the other aldoses. Since the development of this concept has been adequately reviewed elsewhere this introduction will be limited to discussion of the catalysts operating in this type of transformation.

Glucose and its derivatives have served as the principal substances for investigating the catalysis of mutarotation, although the first precise kinetic analysis of the phenomenon was carried out on lactose by Hudson (1) who observed that the reaction is subject to catalysis by both hydroxyl and hydronium ions and who evaluated catalytic constants for these species as indicated in his expression for the pseudo constant: $k = A + B\,[H_3O^+] + C\,[OH^-]$, where A, B and C are constants at a given temperature. Lowry (2) in investigating the mutarotation of tetramethylglucose in anhydrous pyridine and cresol observed that this reaction is

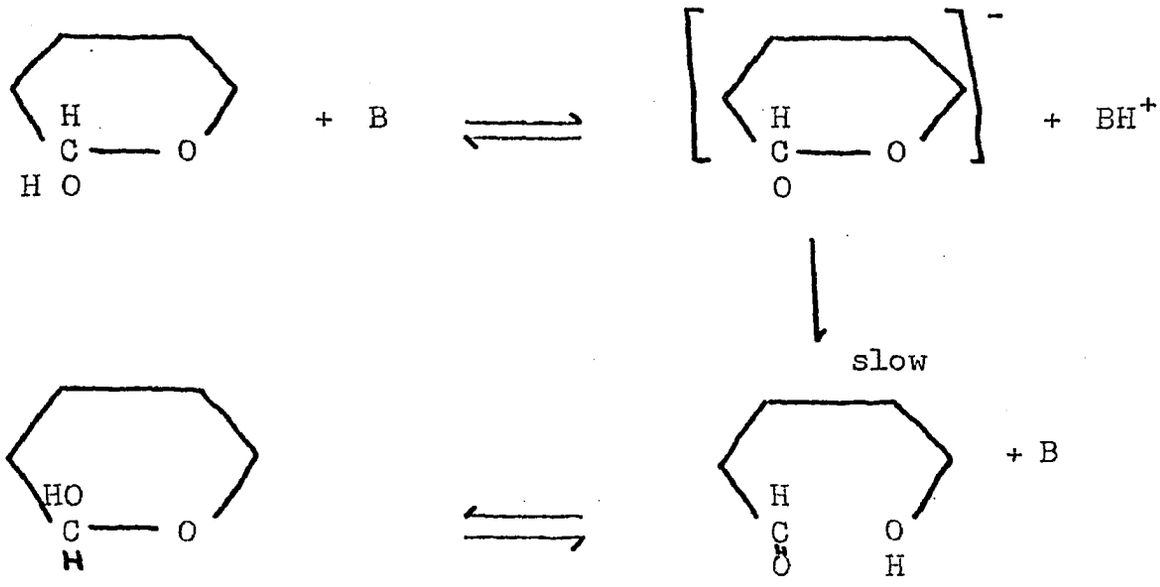
markedly catalyzed by mixtures of these substances and proposed a concerted mechanism for the catalysis involving simultaneous attack on the substrate molecule by the acidic cresol and the basic pyridine. The classical work of J. N. Brönsted (3) on the mutarotation of glucose in aqueous solution greatly amplified that of Lowry in demonstrating that the reaction is subject to catalysis by any proton acceptor or donor.

It remains a curious fact that Brönsted, in his original writing, indicated complete acceptance of the Lowry interpretation, yet the idea of a concerted catalysis gradually fell into disrepute, largely owing to the fact that no third order term appears in Brönsted's rate expression. In 1934 Pederson (4) represented the reaction as occurring by way of a two step mechanism as shown below:

Base catalyzed



Acid catalyzed



The current revival of the concerted mechanism rests principally on the recent work of Swain (5) who in a reinterpretation of Brönsted's data showed that the mutarotation of glucose does indeed permit inclusion of a third order term in the rate expression, and that this term escapes detection because of the magnitude of the water catalysis. In Swain's treatment the concerted mechanism was assumed. This led to expression of the pseudo constant as $k = k_o \left(\sum_N r_n [N]\right)\left(\sum_E r_e [E]\right)$, where $\underline{N}$ is any nucleophilic reagent, $\underline{E}$ any electrophilic reagent, and r_n and r_e are the respective reactivities of these substances with respect to a fixed standard such as water. Application of this expression to the mutarotation of glucose in an acetate buffer and equating Brönsted's experimental coefficients to the appropriate combinations resulted in five simultaneous equations which upon solution gave the following expression:

$$k = 8.8\times10^{-5} + 4.4\times10^{-4}[AcO^-] + 1\times10^{2}[OH^-] + 4\times10^{-5}[HOAc] + 2.4\times10^{-3} + 0.2\times10^{-3}\frac{[AcO^-]}{[HOAc]}$$

This was exactly equivalent to Brönsted's expression except for the presence of the third order term which was too small to be detected experimentally.

More recently Swain (6) has demonstrated the existence of third order kinetics for the mutarotation of tetramethylglucose in pyridine-cresol mixtures (this had not been previously shown by Lowry) and has observed marked catalysis of the reaction by the substances 2-hydroxy-pyridine and 2-hydroxy-4-methylquinoline (7) in which nucleophilic and electrophilic centers are combined within single molecules. This may be regarded in the future as one of the more significant contributions to the general field of homogeneous catalysis as well as to the particular field of enzyme catalyzed reactions.

The status of the catalysis of mutarotation at the beginning of the present work may be summed up as follows:

(a) The most plausible mechanism for catalysis of the mutarotation of either glucose or tetramethylglucose involves a concerted attack on the substrate molecules by nucleophilic and electrophilic centers. These centers may be parts of separate molecules, or ions, or may be incorporated within a single species.

(b) Each of the nucleophilic catalysts previously observed is a proton acceptor (Brönsted base) and each of the electrophilic catalysts is a proton donor (Brönsted acid). Apparently no homogeneous catalysis by any other species was noted in earlier work.

EXPERIMENTAL PART

EXPERIMENTAL PART

Chemicals

The α -D-glucose used in these experiments was prepared at the National Bureau of Standards (lot number 5052). β -D-glucose, D-galactose, D-xylose and L-rhamnose were Pfanstiehl Chemical Company preparations. The β-glucose was recrystallized by Hudson's method (8), and the galactose was purified by the Bureau of Standards procedure (9). Beryllium carbonate was a product of C. A. F. Kahlbaum Chemischfabrik. Ferric perchlorate was obtained from the G. Frederick Smith Chemical Company. All other reagents used in these experiments (calcium carbonate, lithium carbonate, magnesium carbonate, nickelous chloride, cupric chloride, magnesium chloride, calcium chloride, sodium hydroxide, potassium hydrogen phthalate, acetic acid and perchloric acid) were C. P. quality.

Solutions

Standard sodium perchlorate solution was prepared by neutralization of perchloric acid with sodium hydroxide to pH 7.0. The various acetate salts were prepared by addition of standard acetic acid to weighed samples of the carbonates. The concentrations of stock solutions of the hydroscopic salts, or those of uncertain composition, were determined by standard analytical procedures.

Apparatus

A Rudolph polarimeter equipped with a sodium lamp and a thermostated 1-decimeter tube was used for measuring all optical rotations. pH determinations were made with a Beckman pH meter equipped with outside glass and calomel electrodes. A Beckman spectrophotometer (model DU) and 1-centimeter Corex cells were employed for the spectral studies.

Spectral Studies

A 0.2 molar nickelous chloride solution was prepared and its spectral characteristics determined. A maximum in the absorption curve was observed at 395 mu. Optical density readings were made at four wavelengths, 380, 395, 410 and 420 mu, on mixtures prepared by adding x milliliters of the 0.2 molar nickelous chloride solution to (10-x) milliliters of 0.2 molar glucose solution. Optical densities were also determined for solutions of nickelous chloride having the same concentrations of nickelous ion as in the glucose mixtures. This is essentially the method outlined by Job (10) for the investigation of complexes by the method of continuous variations.

Kinetic Method

The rate of mutarotation was followed by the change in optical rotation. The reaction mixtures were prepared as follows: An aliquot of the appropriate salt solution, the ionic strength of which was usually

adjusted by addition of sodium perchlorate, was cooled to the desired temperature (either 18.00 or 18.90°). This was mixed with a weighed sample of the aldose, and, after solution of the sugar, it was diluted to a definite volume. The timer was started at the moment of complete solution of the sugar. The optical rotation was then read at approximately one-hundred-second intervals over a period of three-fourths hour, and the equilibrium rotational value was observed after twenty-four hours. Values for the observed rate constants in these experiments were obtained in the usual manner from the following expression:

$$k = k_1 + k_2 = 1/t \log r_o - r_{oo} / r_t - r_{oo}$$

The results of a typical velocity determination are shown in Table I and Figure 1. All velocity data are expressed in decadic logarithms, with the second as the unit of time.

TABLE I

TYPICAL VELOCITY DETERMINATION
(Mutarotation of 0.5 M aqueous glucose solution. pH= 5.8; t= 18.00°.)

Seconds	Reading	$R_t - R$	$\log(R_t - R)$
67	9.580	4.820	.68305
139	9.548	4.789	.68015
207	9.471	4.711	.67311
310	9.350	4.590	.66181
409	9.274	4.514	.65456
524	9.174	4.414	.64483
632	9.072	4.312	.63468
751	8.963	4.203	.62356
895	8.840	4.080	.61066
1019	8.732	3.972	.59901
1129	8.654	3.894	.59040
1318	8.491	3.731	.57183
1453	8.407	3.647	.56194
1637	8.261	3.501	.54419
1814	8.137	3.377	.52853
2044	7.995	3.235	.50987
2211	7.876	3.116	.49360
2412	7.752	2.992	.47596
2841	7.520	2.760	.44091
3054	7.383	2.623	.41880
Equilibrium	4.760		

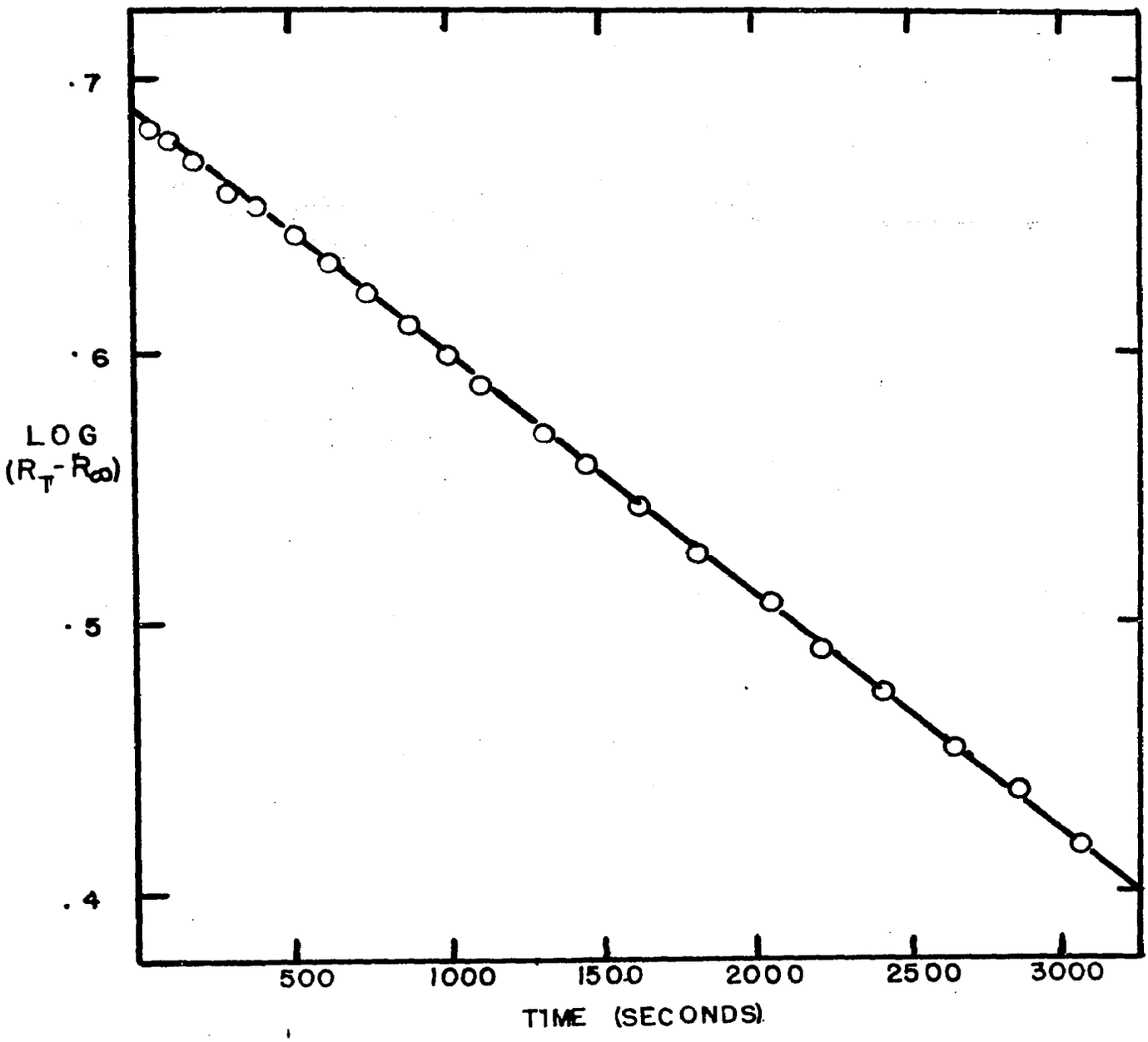


Figure 1. Typical Kinetic Run. Mutarotation of 0.5M Glucose in a Water Solution. ($t = 18.00^{\circ}$ C pH = 5.8)

RESULTS AND DISCUSSION

RESULTS AND DISCUSSION

Preliminary studies of the effect of metallic ions on the observed rate of mutarotation of glucose in aqueous medium showed that there was a marked increase in the pseudo first order rate constant. The observed increase could not be attributed to either a primary or a secondary salt effect since 0.2 was the maximum ionic strength used in these experiments, and Brönsted (3) showed that the reaction was independent of ionic strength up to this value. It must also be concluded that the variation in k is not due to complexing of glucose by the metal ion for if a complex were formed in appreciable concentration, a shift in the equilibrium rotational readings would have been observed. The absence of such a shift in any of the systems studied is shown in Table II. Further verification that a complex is not responsible for the effect was obtained in the experiment where the metal ion concentration was held constant and that of the glucose varied. These results are indicated in Table III. From the fact that the observed k's remained constant, it is evident that a stable complex either is not formed or, if it is formed, is present in insufficient concentration to account for the observed phenomena. Application of the method of continuous variation to the nickel glucose system (see Table IV, Figure 2) did indicate the formation of a one to one complex, however, the small change in the optical density substantiates the previous conclusion that the concentration of complex is not large enough to be responsible for the increase in k.

TABLE II

COMPARISON OF EQUILIBRIUM ROTATIONAL READINGS IN THE PRESENCE AND ABSENCE OF METALLIC IONS (GLUCOSE = 0.5M)

Metal Ion	Molarity	Equilibrium Reading*
None		4.778
Lithium	0.050	4.768
Beryllium	0.050	4.771
Magnesium	0.050	4.762
Calcium	0.050	4.778
Copper	0.050	4.781
Iron	0.0467	4.765
Nickel	0.050	4.742

* These readings represent the average value of several velocity determinations.

TABLE III

EXPERIMENT TO DETERMINE THE EFFECT OF VARYING THE GLUCOSE CONCENTRATION ON THE PSEUDO CONSTANT

Cation	Molarity Ion	Molarity Glucose	k x 10^3 x sec^{-1}
* Mg^{++}	0.05	0.3	1.52
		0.4	1.53
		0.5	1.52
** Cu^{++}	0.04	0.3	1.92
		0.5	1.94

* Experiments were run in a 0.1M acetate and 0.1 acetic acid, (u = 0.2, t = 18.90° C)

** Experiments were run in a water solution and the (t = 18.00° C)

TABLE IV

RESULTS OF CONTINUOUS VARIATION STUDY ON THE NICKEL GLUCOSE COMPLEX

Volume Ni^{++} Solution, ml.	Wave Length, mu											
	380			395			410			420		
	Optical Density		Difference	Optical Density		Difference	Optical Density		Difference	Optical Density		Difference
	Ni^{++} Soln*	Mixture**		Ni^{++} Soln*	Mixture**		Ni^{++} Soln*	Mixture**		Ni^{++} Soln*	Mixture**	
1	.079	.068	.011	.105	.087	.018	.079	.065	.014	.049	.041	.008
2	.160	.133	.027	.212	.181	.031	.156	.126	.030	.103	.083	.020
3	.250	.212	.038	.329	.284	.045	.251	.210	.041	.162	.132	.030
4	.348	.299	.049	.459	.394	.065	.349	.291	.058	.230	.194	.036
5	.429	.367	.062	.560	.472	.078	.434	.362	.072	.283	.236	.047
6	.488	.439	.049	.648	.585	.063	.494	.437	.057	.318	.250	.038
7	.578	.545	.033	.754	.706	.048	.575	.534	.041	.379	.354	.025
8	.655	.630	.025	.859	.829	.030	.660	.633	.027	.434	.418	.016
9	.814	.805	.009	1.00	.850	.015	.812	.797	.015	.542	.533	.009

* Indicated volume of 0.2 M nickel solution made up to final volume of 10 ml. with water.

** Indicated volume of 0.2 M nickel solution made up to final volume of 10 ml. with 0.2 M glucose solution.

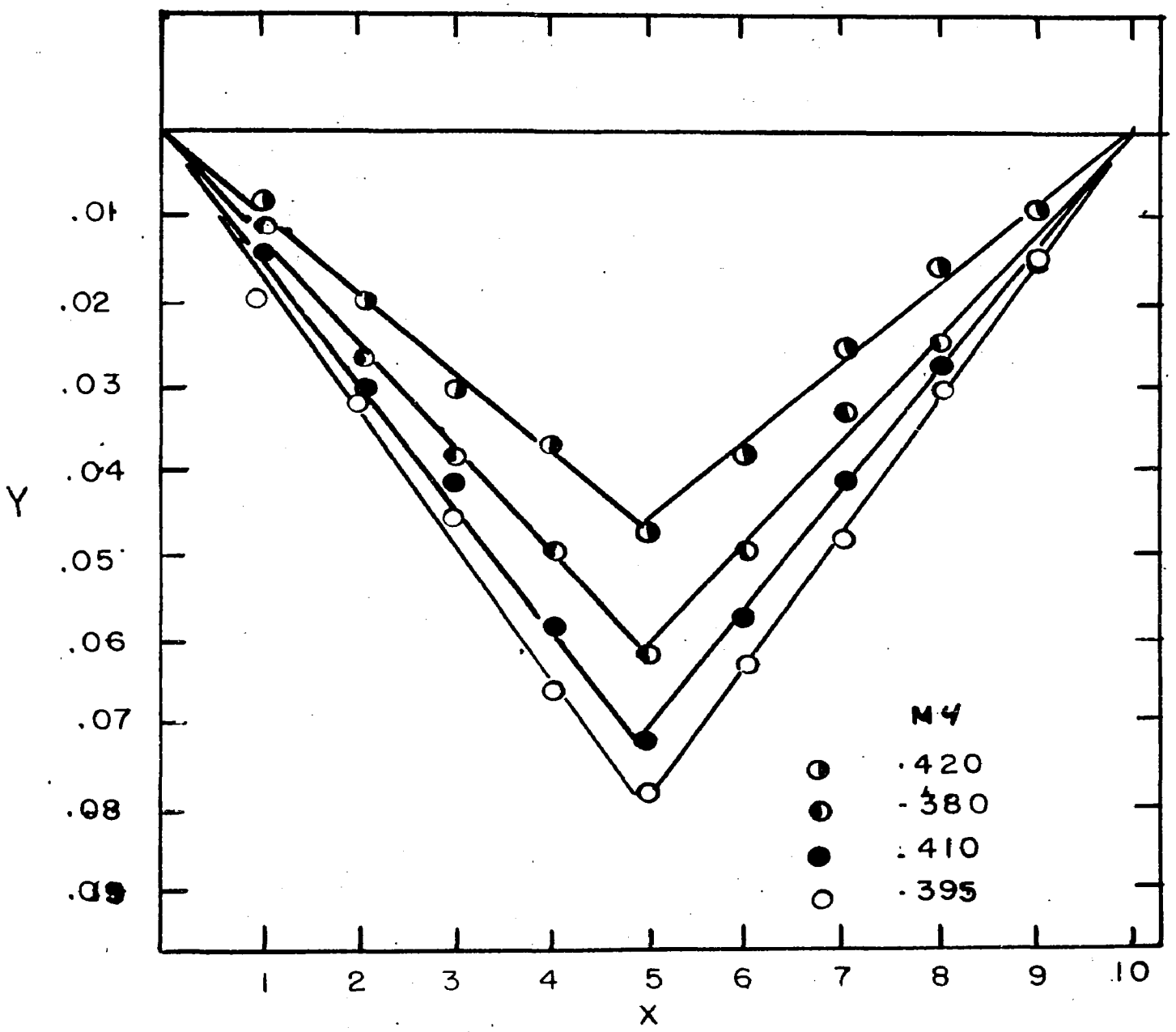


Figure 2. Results of Continuous Variation Study on Nickel Glucose Solutions.
x = ml. of 0.2M Ni^{++} solution made up to 10 ml.
Y = (Optical Density of unreacted Nickel)
-(Optical Density of reacted Nickel)

The effect of the metal ions on k, therefore, must be attributed to a catalytic action, and it should be possible to evaluate catalytic constants for the various cations. By maintaining the pH of the medium between 4 and 6 the pseudo rate constant in water solutions may be represented by the following expression:

$$k = k_o + k_m [M^+]$$

where k_o is the water catalyzed constant and k_m is the catalytic constant for the particular metal ion. In obtaining catalytic constants for magnesium, calcium and cupric ions in the mutarotation of glucose the reaction was carried out at various concentrations of the ions and the pseudo constants determined. These results are given in Table V. Plots of k versus concentration are shown in Figure 3.

Catalytic constants for lithium, beryllium and ferric ions were evaluated from experiments in perchloric acid solution by application of the expression

$$k = k_o + k_{H_3O} [H_3O^+] + k_m \quad [M^+]$$

in which k is again the observed pseudo constant. The catalytic constant for hydronium ion was rechecked with perchloric acid solutions and found to be 2.43×10^{-3} at 18°C in close agreement with Brönsted's value of 2.41×10^{-3} (3). (In plotting hydronium ion concentration against pseudo constant (Figure 4) stoichiometric concentrations of perchloric acid were used for the concentrations of hydronium ion). The results of the velocity determinations with these metal ions are given in Table VI. Data for magnesium ion obtained in perchloric acid are also included in this tabulation, and it will be noticed that the

TABLE V

EFFECT OF CALCIUM MAGNESIUM AND CUPRIC IONS IN THE MUTAROTATION OF GLUCOSE IN WATER SOLUTIONS
(pH between 4 and 6, t = 18.00°C, Glucose = 0.5M)

Metal Ion	Concentration x 10^2 moles	k x 10^4 sec^{-1}	*Catalytic Constant x 10^4
Mg^{++}	2.00	.924	1.72
	4.00	.945	
	5.00	.970	
	7.00	1.00	
Ca^{++}	2.50	.950	4.40
	5.00	1.12	
Cu^{++}	2.00	.985	11.9
	2.50	1.35	
	4.00	1.95	
	5.00	2.25	
None		.880	

* Catalytic constants calculated from the slopes in Figure 3.

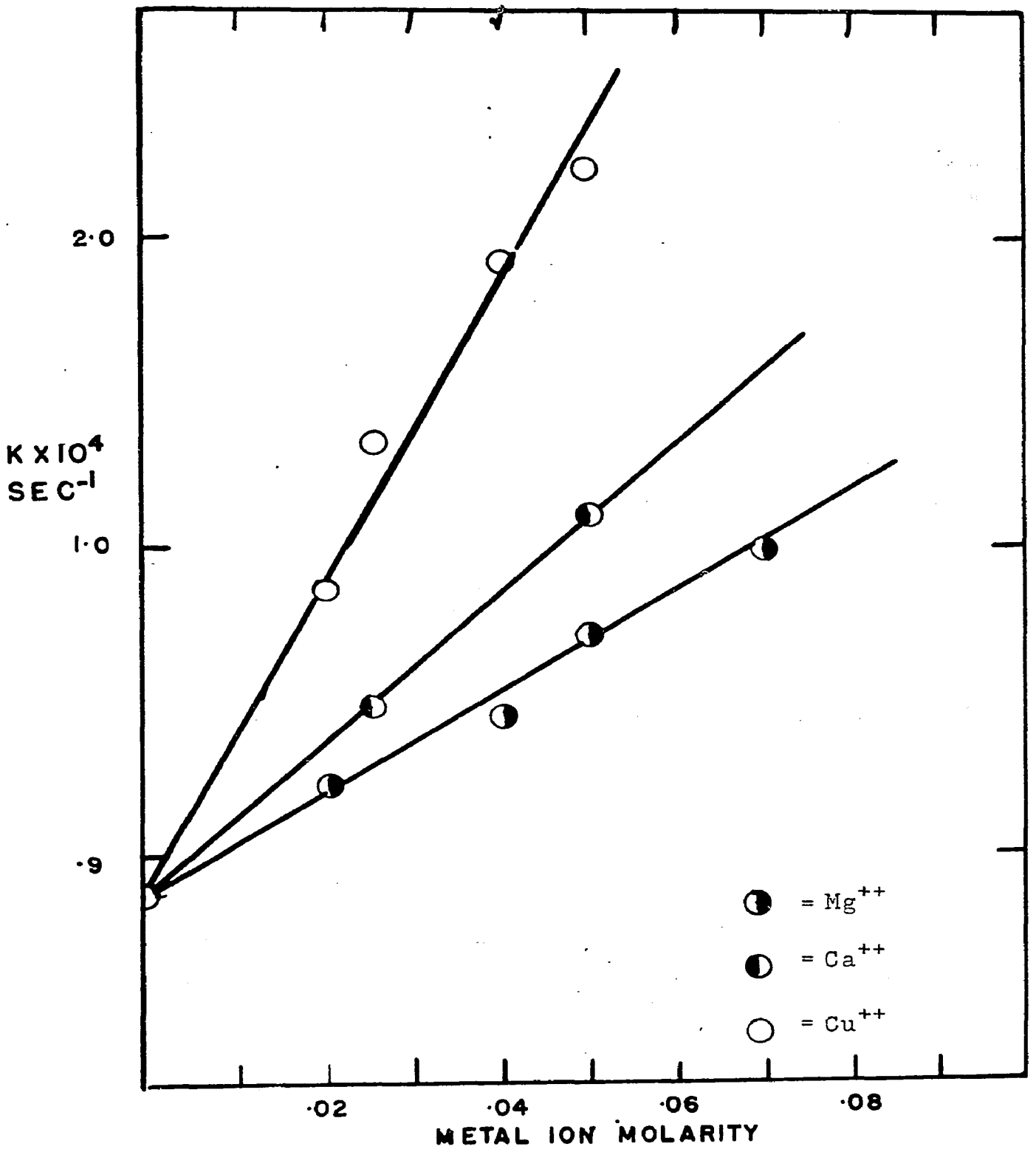


Figure 3. Relation of Pseudo Constant (k) to Metal Ion Concentration.

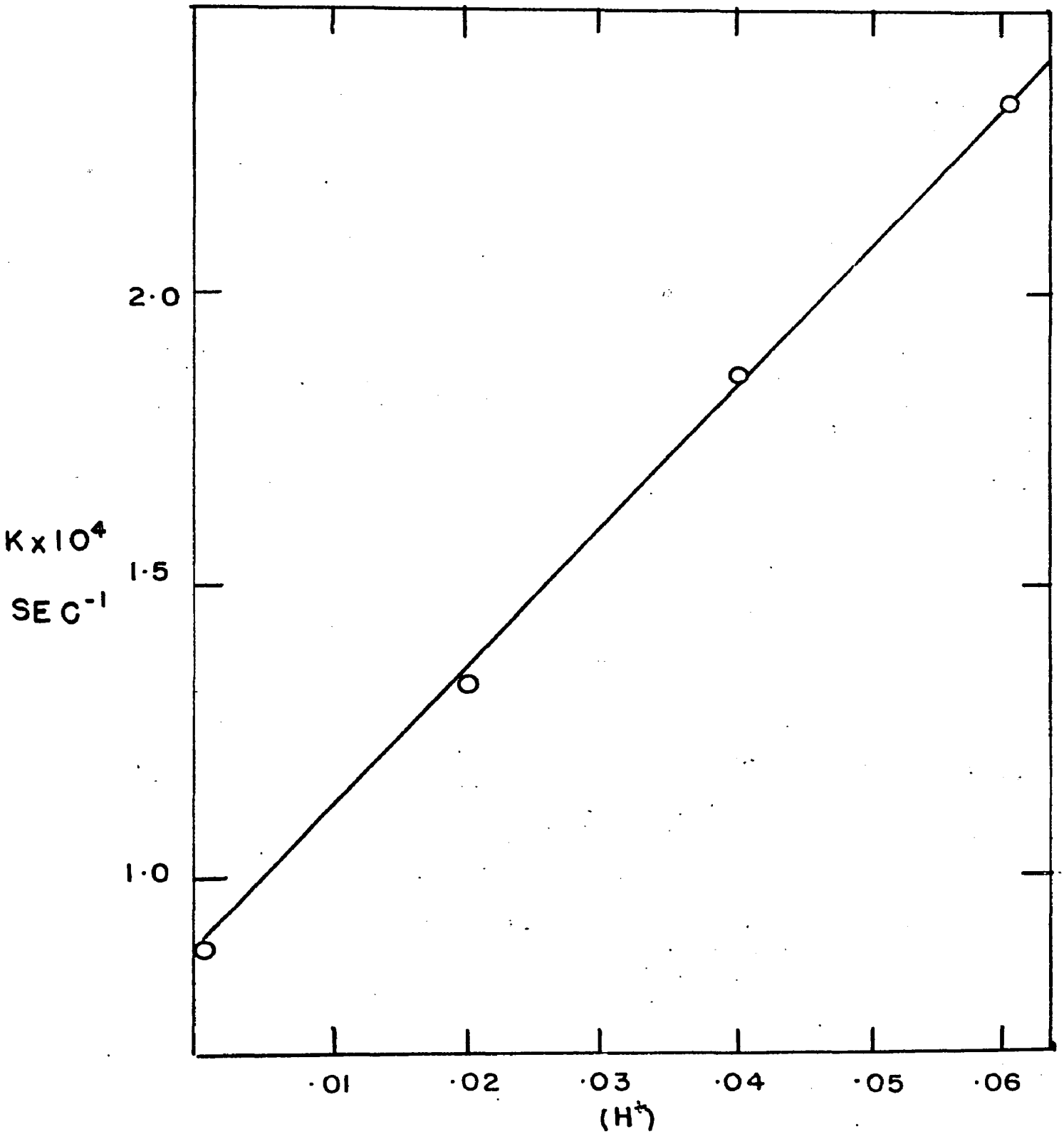


Figure 4. Relation of Pseudo Constant (k) to Hydrogen Ion Concentration.

TABLE VI

EFFECT OF FERRIC, MAGNESIUM, CALCIUM, BERYLLIUM AND LITHIUM IONS IN PERCHLORIC ACID MEDIUM IN MUTAROTATION OF GLUCOSE
(t = 18.00°C, Glucose = 0.5M)

Cation	Molarity Ion	Molarity of $[H^+]$	k x 10^4 x sec^{-1}	Catalytic Constant x 10^5
*Li^+	0.0500	0.0128	1.21	3.00
	0.0500	0.0224	1.44	3.30
	0.100	0.0230	1.47	3.00
Mg^{++}	0.0228	0.0640	2.47	17.5
	0.0250	0.0273	1.58	17.2
	0.0500	0.0227	1.60	17.5
	0.0500	0.0190	1.43	17.6
Be^{++}	0.0250	0.0223	1.51	18.0
	0.0500	0.0223	1.50	17.9
	0.0500	0.0223	1.46	17.8
**Fe^{++}	0.0155	0.0640	2.55	100
	0.0234	0.0640	2.66	
	0.0467	0.0640	2.90	

* Catalytic constants calculated from the following expression

$$k_m = \frac{k(\text{observed}) - 8.80 \times 10^{-5} - 2.43 \times 10^{-3} (H^+)}{(\text{Metal})}$$

** Catalytic constant calculated from the slope of the line in Figure 5.

acidity of the medium does not affect the magnitude of its catalytic constant. Figure 5 shows a plot of ferric ion concentration against pseudo constant at constant hydronium ion concentration (0.0640M) for which the slope of the line equals the catalytic constant for the former species.

The catalytic constants for acetate ion and acetic acid were re-checked before attempting to evaluate the effect of metal ions on the reaction in an acetate buffer. Since mutarotation is independent of hydronium ion between pH 4 and pH 6 the constants may be determined by varying one species and holding the other constant. The pseudo constant is given by the following expression

$$k = k_o + k_{Ac}(AcO^-) + k_{HAc}(HOAc)$$

These results are shown in Table VII and Figure 6. The values of 4.46×10^{-4} for acetate ion and 4.20×10^{-5} for acetic acid agree closely with Brönsted's values of 4.45×10^{-4} and 4.0×10^{-5} respectively (3).

The effect of metallic ions on the observed rate of mutarotation in an acetate buffer indicated an initial decrease in the pseudo constant with increasing metal ion concentration. There was a similar effect with xylose, rhamnose and galactose, demonstrating that the phenomena was not specific for the glucose molecule. The same rate constant was observed for -glucose as for the -isomer in the presence of magnesium ion indicating a lack of preference of the cation for one stereoisomer over the other. These results are tabulated in Tables VIII, IX, X. The effect of lithium and magnesium ions is also

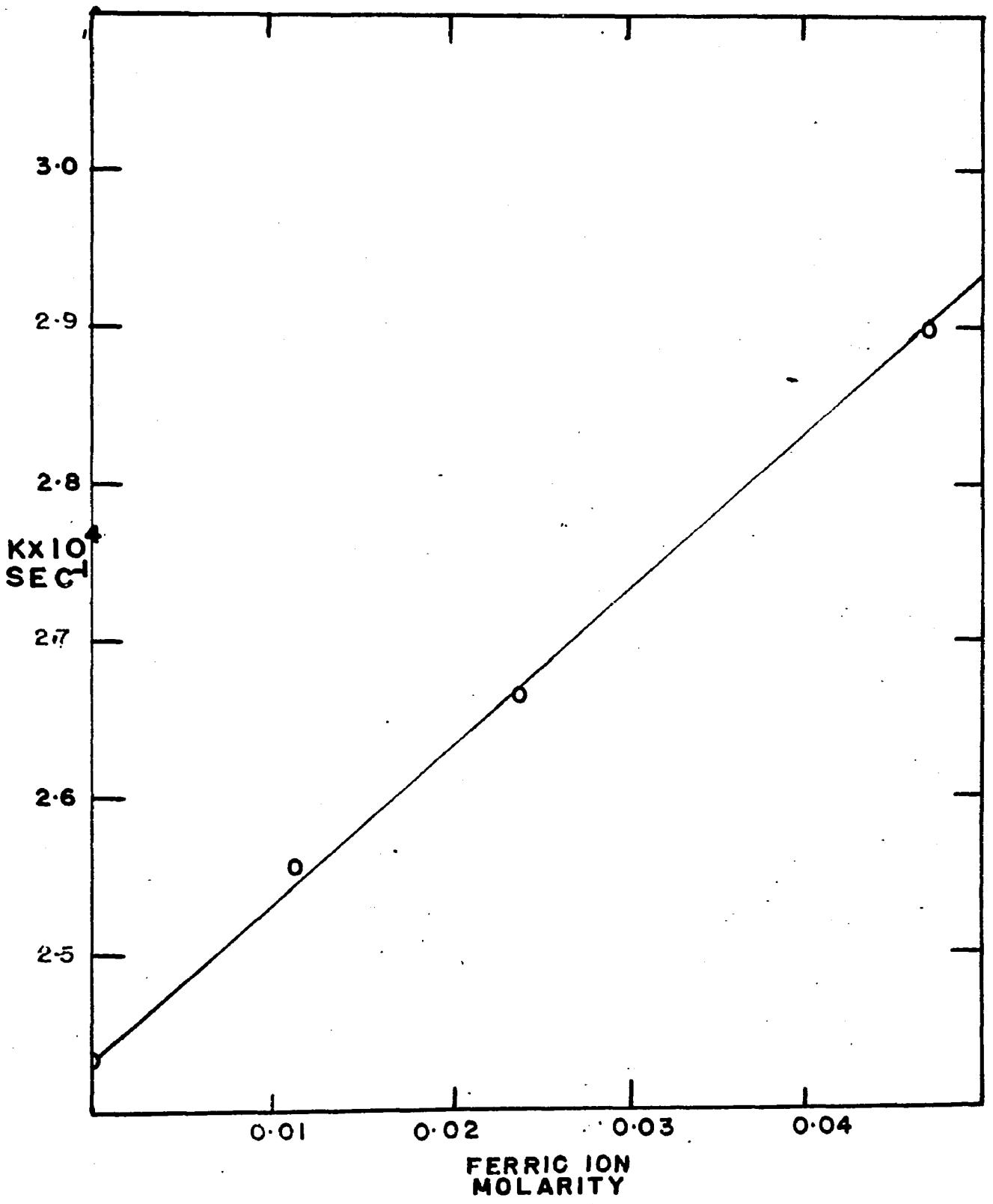


Figure 5. Relation of Pseudo Constant (k) to Ferric Ion Concentration.

TABLE VII

EXPERIMENTS TO DETERMINE THE CATALYTIC CONSTANTS OF ACETATE ION AND ACETIC ACID ON THE MUTAROTATION OF GLUCOSE
(t = 18.00°C, = 0.2, Glucose = 0.5M)

Molarity of Acetate	Molarity of Acetic Acid	k x 10^3 x sec^{-1} Observed	*k x 10^3 x sec^{-1} Calculated
0.100	0.100	1.38	1.37
0.150	0.100	1.61	1.59
0.200	0.100	1.81	1.80
0.100	0.100	1.38	1.37
0.100	0.150	1.39	1.39
0.100	0.200	1.42	1.41

* This was calculated from the following expression:

$$k = 8.80 \times 10^{-5} + 4.20 \times 10^{-5} (HOAc) + 4.46 \times 10^{-4} (AcO^-)$$

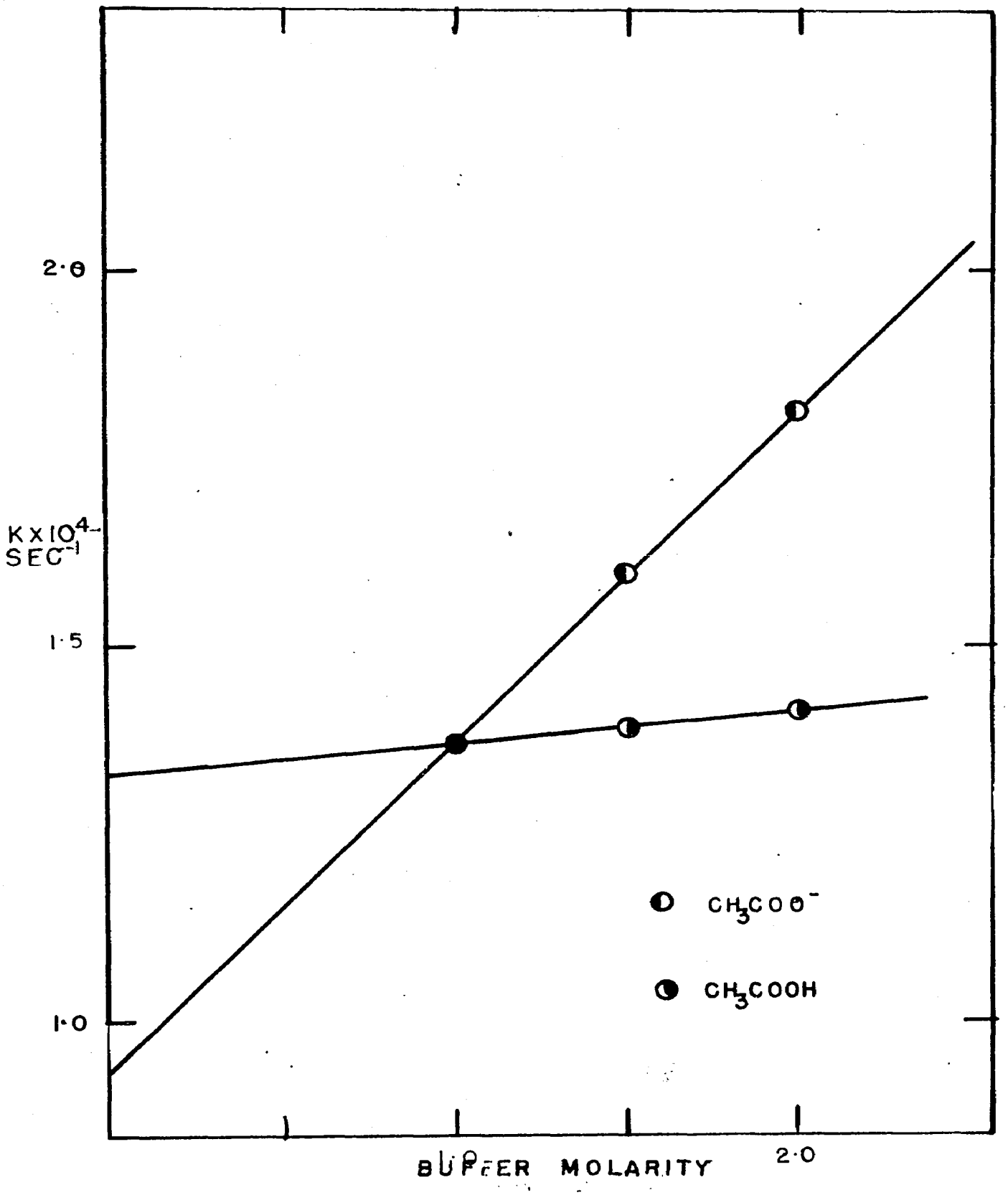


Figure 6. Relation of Pseudo Constant (k) to Acetate Ion and Acetic Acid Concentration.

TABLE VIII

EXPERIMENTS TO DETERMINE THE EFFECT OF METALLIC IONS ON THE MUTAROTATION OF GLUCOSE IN ACETATE BUFFER.
($t = 18.90^{\circ}C$, $u = 0.2$, $HOA = 0.1M$, $AcO^{-} = 0.1M$, Glucose $= 0.5M$)

Experiment	Metal Ion	Molarity x 10^2	k x 10^4 sec^{-1}
5,9,13,20,22	None		1.52
*12	None		1.52
17	Mg^{++}	1.50	1.47
15		2.50	1.44
16		3.50	1.42
7,8,10,23		5.00	1.38
19,42	Ca^{++}	5.00	1.44
60	Li^{+}	2.50	1.40
40,55		5.00	1.34
61		7.50	1.37
41,58		10.0	1.42
36	Be^{++}	2.50	1.39
35,37		5.00	1.30
33,39	Ni^{++}	2.50	1.40
34,38		5.00	1.30
**66,75	None		1.86
68	Mg^{++}	2.50	1.79
64,67		5.00	1.75
78		7.50	1.80
63,71		10.0	1.87

* Reaction run in the absence of sodium perchlorate.

** The remainder of these experiments were run in 0.2M acetate and 0.2M acetic acid (u = 0.4).

TABLE X

EXPERIMENTS TO DETERMINE THE EFFECT OF LITHIUM AND MAGNESIUM IONS ON THE MUTAROTATION OF GLUCOSE ACETATE BUFFER
($t = 18.00°C$, $u = 0.2$, $HOAc = 0.1M$, $AcO^- = 0.1M$, Glucose $= 0.5M$)

Metal Ion	Molarity x 10^2	k x 10^4 sec^{-1}
Li^+	2.00	1.34
	4.00	1.31
	5.00	1.30
	6.00	1.31
	8.00	1.33
	10.00	1.37
Mg^{++}	5.00	1.30
None		1.38

shown in Figure 7. Experiment 12 (Table VIII) was run in the acetate buffer minus the sodium perchlorate and constitutes a verification of Brönsted's observation of the absence of primary and secondary salt effects.

The results of the experiments with acetate buffers indicate that at the lower metal ion concentration a reaction occurs which masks the effect of the catalysis by the metal ion. A logical explanation of such a phenomenon is that the concentration of one or more of the catalytic species present is being effectively lowered. This conclusion is in direct accord with R. P. Bell's work on the incomplete dissociation of salts (11,12,13). Bell presents data for the decomposition of nitramide (13), a general base catalyzed reaction, in the presence of calcium and barium salts of carboxylic acids. The observed velocity constants were smaller in solutions of the salts of these bivalent cations than they were in solutions of the corresponding sodium salts. In explaining these results it was assumed that the calcium and barium salts are incompletely dissociated, and the respective dissociation constants were calculated on this basis. In any reaction which is subject to catalysis by both acids and bases the problem of evaluating such dissociation constants is more complex. The rate expression will contain factors representing the reaction between the transition state and the oppositely charged species present, and it becomes difficult to distinguish between individual reactions. Thus Bell's work, while it affords a reasonable explanation for the observed phenomena, it does not provide a satisfactory method for computing the dissociation constants.

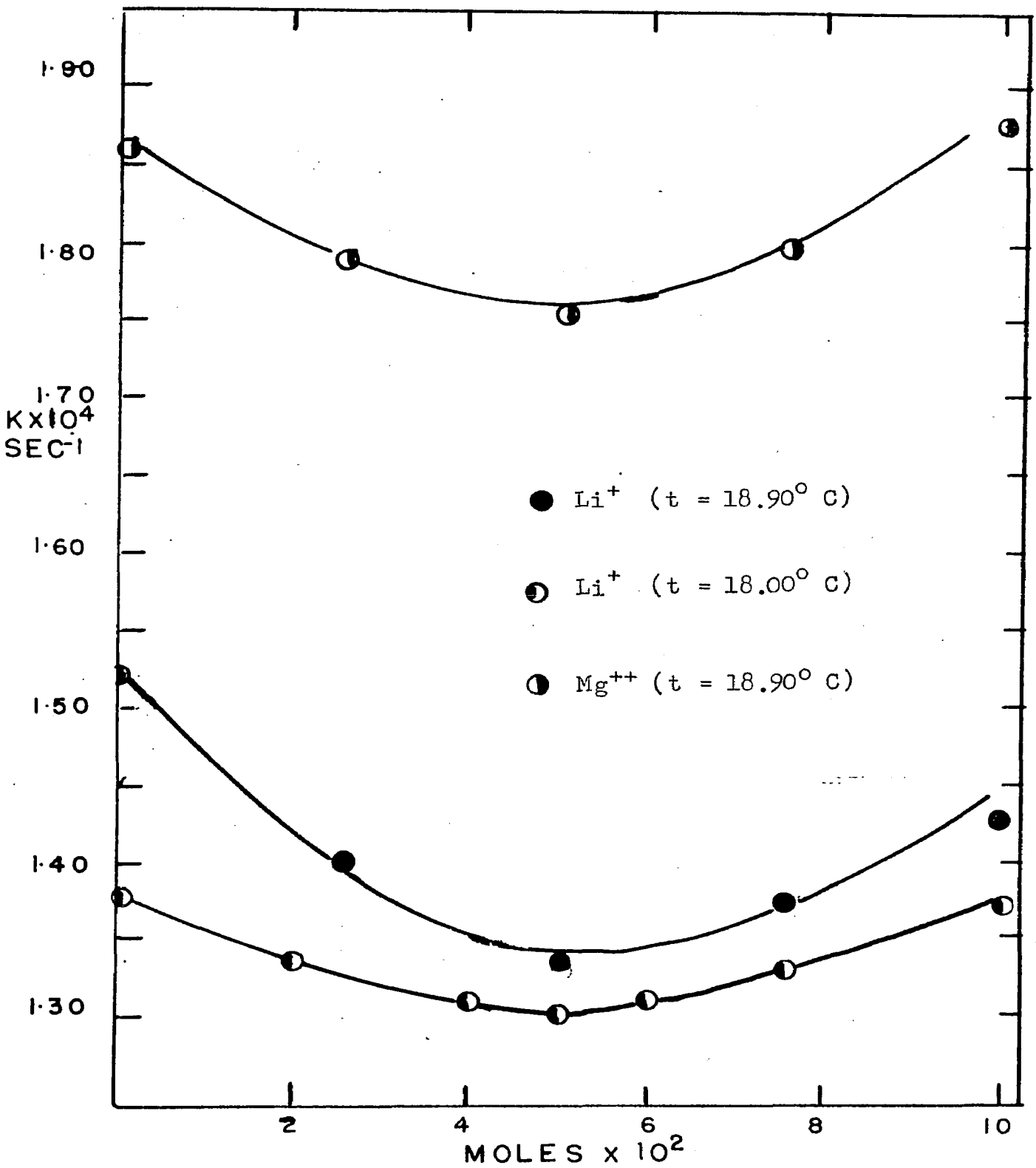


Figure 7. Relation of Mg^{++} and Li^{+} Molarity in Acetate Buffer to Pseudo Constant (k).

The only case which lends itself to a reasonable mathematical analysis is the lithium acetate system. The bi- and trivalent metallic ion systems involve too many variables in the form of catalytic species (such as $[MeAcO]^+$) to be handled quantitatively.

In analyzing the lithium acetate system two methods, A and B, will be discussed. The assumptions underlying method A are as follows:

1) Lithium acetate is incompletely dissociated.

2) The observed rate constant may be expressed as follows:

$$k = k_o + k_1 (HOAc) + k_2(AcO^-) + k_3 (Li^+)$$

The dissociation constant K_d for lithium acetate will be represented by the following equations:

$$LiAc \rightleftharpoons Li^+ + AcO^- \qquad (1)$$

$$\frac{(Li^+)\ (AcO^-)}{(LiOAc)} = K_d \qquad (2)$$

Let x = the original lithium ion concentration

a = the original acetate ion concentration

y = the amount of undissociated lithium acetate.

Therefore $K_d = \frac{(x-y)(a-y)}{y}$

Solving this equation for y yields the following:

$$y = \frac{a + x + K_d - [(a + x + K_d)^2 - 4ax]^{1/2}}{2} \qquad (3)$$

The observed velocity constant is given by the following:

$$k = k_o + k_1 (HOAc) + k_2(a-y) + k_3(x-y) \qquad (4)$$

Substituting equation 3 into 4 yields:

$$k = k_o + k_1(HOAc) + 1/2k_2 \{(a-x-K_d)+[(a+x+K_d)^2-4ax)]^{1/2}\} \qquad (5)$$
$$1/2k_3\{x-a-K_d + [(a+x+K_d)^2 - 4ax]^{1/2}\}$$

This gives an expression in which the observed rate constant is a function of the metal ion concentration. If this function is in accord with the facts the first derivative should be zero and the second derivative should be greater than zero if equation (5) possesses a minimum point. Carrying out the required differentiation gives the following equations:

$$\frac{dk}{dx} = -1/2k_2 + 1/2k_2 \left[\frac{(x-a+K_d)}{[(a+x+K_d)^2-4ax)^{1/2}}\right] + 1/2k_3 + 1/2k_3 \left[\frac{(x-a+K_d)}{(a+x+K_d)^2-4ax)^{1/2}}\right] \quad (6)$$

$$\frac{d^2k}{dx^2} = \frac{k_2\,[(a+x+K_d)^2 - 4ax)] - k_3(x-a+K_d)^2}{2\,[(a+x+K_d)^2 - 4ax]^{3/2}}$$

$$+ k_3[(a+x+K_d)^2 - 4ax] - \frac{k_3(x+K_d-a)^2}{2\,[(a+x+K_d)^2 - 4ax]^{3/2}} \quad (7)$$

In order that $\frac{d^2k}{dx^2} > 0$ the necessary and sufficient condition is that:

$$(a+x+K_d)^2 - 4ax > (x-a+K_d)^2$$

On expanding this expression, it is seen that the left side is greater than the right, and the left side of the inequality sign will be positive if $a^2 + K_d^2 + x^2 + 2aK_d + 2xK_d > 2ax$. Substituting $\underline{a} = 0.1$ and $\underline{x} = 0.05$ the values that these variables assume at the minimum point, in the above expression yields; $K^2_d + 0.3K_d + 0.0125 > 0.01$ which is obviously true for any value of $\underline{K_d}$. Therefore, the function does have a minimum point. To calculate $\underline{K_d}$ the first derivative in equation (6) is set equal to zero, and the values of the variables at the minimum are

substituted into the expression and K_d is then obtained by approximation. This gives a value of 1.4 for the dissociation constant of lithium acetate. Employing this value, and performing the necessary calculations the calculated rate constants listed in Table XI are obtained.

In method B, the original assumption regarding the dissociation constant for lithium acetate will be retained, in addition it will be assumed that the catalysis follows third order kinetics. With this new assumption, k' may be expressed as follows:

$$k = k_o + k_1 (HOAc) + k_2(AcO^-) + k_3(Li^+) + k_4(Li^+)(AcO^-) \qquad (9)$$

Combining $k - k_o - k_1(HOAc)$ into one constant K and substituting the various algebraic expressions for the lithium and acetate ions yields the following equation:

$$\begin{aligned} K &= 1/2k_2 [a - x - K_d + [(a + K_d + x)^2 - 4ax]^{1/2}] \\ &+ 1/2k_3 [x - a - K_d + [(a + K_d + x)^2 - 4ax]^{1/2}] \\ &+ k_4 K_d [K_d + a + x - [(a + K_d + x)^2 - 4ax]^{1/2}] \end{aligned} \qquad (10)$$

which on differentiation gives:

$$\frac{dK}{dx} = -1/2k_2 + 1/2k_2 \left[\frac{x-a+K_d}{(a+K_d+x)^2 - 4ax)^{1/2}}\right] + 1/2k_3 + 1/2k_3 \left[\frac{x-a+K_d}{[(a+K_d+x)^2 - 4ax]^{1/2}}\right] \qquad (11)$$

$$+ k_4K_d - k_4K_d \left[\frac{x - a + K_d}{[(a + K_d + x)^2 - 4ax]^{1/2}}\right]$$

at the minimum point $\frac{dK}{dx} = 0$ and

$$k_4 = \frac{1/2k_2 - 1/2k_3 - \left[\frac{x - a + K_d}{[(a + K_d + x)^2 - 4ax]^{1/2}}\right](1/2k_2 + 1/2k_3)}{K_d \left[1 - \frac{x - a + K_d}{[(a + K_d + x)^2 - 4ax]^{1/2}}\right]} \qquad (12)$$

TABLE XI

COMPARISON OF EXPERIMENTAL VALUES FOR k WITH THOSE DERIVED BY METHODS A AND B

Molarity of Lithium Ion	Experimental $k \times 10^4 \times sec^{-1}$	Calculated $k \times 10^4 \times sec^{-1}$ A	B
0.0200	1.34	1.37	1.36
0.0400	1.32	1.36	1.35
0.0500	1.30	1.36	1.34
0.0600	1.32	1.36	1.345
0.0800	1.33	1.37	1.35
0.100	1.37	1.37	1.37
*0.200	1.74	1.80	1.75

* Reaction mixture was 0.2M in Acetic Acid and 0.2M in Acetate Ion (u = 0.2)

Substituting this expression for k_4 into equation (10), gives an equation in K_d which is capable of solution and yields, by approximation 0.55 as the dissociation constant. If this value along with the other known variables are substituted into equation (12) it gives 2.0×10^{-4} as the third order catalytic constant. Pseudo constants calculated from equation (9) at different lithium ion concentrations, using the above values for K_d and k_4, are shown in Table XI.

The results obtained by means of these two methods show that method B, while it does not allow complete agreement with the observed effect, is an improvement over method A where the third order term was neglected.

Application of Swain's analysis (5) to these results permits the following calculation. As shown previously the pseudo constant k may be expressed as follows:

$$k = v/[c] = k_o \left(\sum_N r_N[N] \right)\left(\sum_E r_E [E] \right) \qquad (13)$$

Both r_N and r_E will be set equal to 1.00 for water.

The experimental expression for the rate is:

$$k = 8.80 \times 10^{-5} + 4.46 \times 10^{-4} (AcO^-) + 4.2 \times 10^{-5} (HOAc) + 3.0 \times 10^{-5}(Li^+) \qquad (14)$$

This expression contains second order terms, but no third order terms. To calculate the magnitude of the third order terms, equation (13) is applied to this experimental expression with the following results:

$$k = k_o([H_2O] + r_{AcO^-}[AcO^-])([H_2O] + r_{HOAc}[HOAc] + r_{Li^+}[Li^+]) \qquad (15)$$

This yields six cross combinations of nucleophilic and electrophilic reagents which must be considered. These are shown in Table XII.

TABLE XII

COMBINATIONS OF NUCLEOPHILIC AND ELECTROPHILIC REAGENTS CONTRIBUTING TO EACH KINETIC ORDER

	$k_o r_N[N] r_E[E]$	Expected Kinetic Order
1.	$k_o[H_2O]^2$	K_1
2.	$k_o r_{Ac^-}[AcO^-][H_2O]$	$K_2[AcO^-]$
3.	$k_o[H_2O] r_{HOAc}[HOAc]$	$K_3[HOAc]$
4.	$k_o r_{AcO^-}[AcO^-] r_{HOAc}[HOAc]$	$K_4[AcO^-][HOAc]$
5.	$k_o[H_2O] r_{Li^+}[Li^+]$	$K_5[Li^+]$
6.	$k_o r_{AcO^-}[AcO^-] r_{Li^+}[Li^+]$	$K_6[Li^+][AcO^-]$

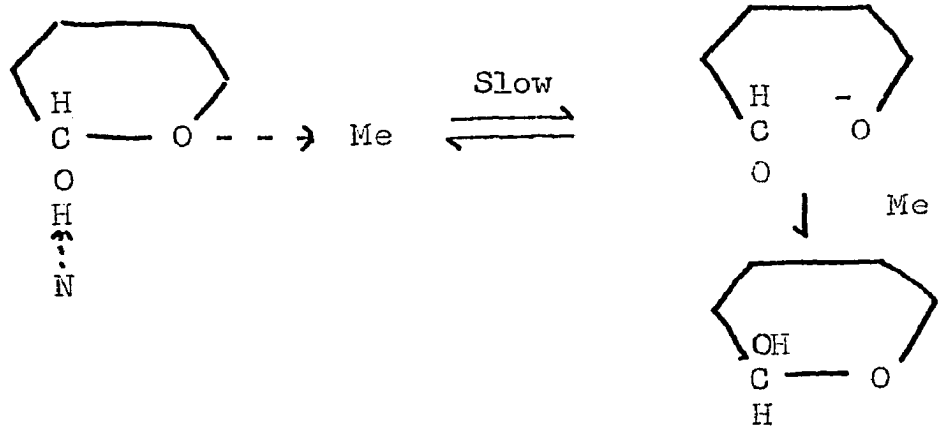
The experimental coefficient for the uncatalysed term, 8.80×10^{-5}, must be set equal to k_1, the experimental coefficient for the acetate term, 4.46×10^{-4}, must be set equal to k_2; k_3 equals 4.20×10^{-5}; and k_5 equals 3.00×10^{-5}. These equations are capable of solution and give the following expressions:

$$k = 2.90\times10^{-8}([H_2O]+2.80\times10^{2}[AcO^-])([H_2O]+2.60\times10^{1}[HOAc]+1.88\times10^{1}[Li^+]$$

$$k = 8.75\times10^{-5}+4.46\times10^{-4}[AcO^-]+4.15\times10^{-5}[HOAc^-]+3.00\times10^{-5}[Li^+]$$
$$+2.10\times10^{-4}[Ac^-][HOAc]+1.53\times10^{-4}[Li^+][HOAc]$$

The second of these calculated expressions agrees exactly with the original experimental expression, except for the presence of the final third order terms. The third order constant for the lithium ion - acetic acid term agrees fairly well with the one derived previously.

In recapitulation it should be stated that this is the first time that metallic ions have been shown to act as electrophilic reagents in the catalysis of mutarotation. Moreover, in view of the above analysis of the lithium acetate data, it appears probable that this catalysis proceeds by way of a concerted mechanism. This mechanism, in which a simultaneous attack by the metal ion and the nucleophilic reagent occurs in the rate determining step, is shown below.



SUMMARY

SUMMARY

1. It was observed that metallic ions catalyze the mutarotation of glucose. This is the first time that metal ions have been known to act as electrophilic reagents in the catalysis of this particular reaction.

2. Catalytic constants for lithium, beryllium, magnesium, calcium, cupric and ferric ion were evaluated at 18.00° C.

3. A diminution in the pseudo first order constant was observed for the mutarotation of glucose, galactose, rhamnose, and xylose in acetate buffer and in the presence of metal ions. This effect was attributed to the incomplete dissociation of the acetate salts.

4. Assuming incomplete dissociation of the acetate salts, it was possible to analyse the lithium acetate system by two distinct methods:

   A. Assumes that the reaction follows second order kinetics.

   B. Assumes that third order kinetics apply to the metal ion catalysis of mutarotation. While the calculated values for the pseudo constant by method B do not agree completely with the experimental values, they do show a marked improvement over method A.

5. Since third order kinetics may be applied to this reaction a mechanism is postulated in which the metal ion and nucleophilic reagent attack the aldose simultaneously in the rate determining step.

REFERENCES

REFERENCES

1. Hudson, C. S., Z. physik Chem., 44, 487 (1903).
2. Lowry, T. M., Faulkner, I. J., J. Chem. Soc., 127, 2883 (1925).
3. Brönsted, J. N., Guggenheim, E. A., J. Am. Chem. Soc., 49, 2554 (1927).
4. Pederson, K. J., J. Phys. Chem., 38, 581 (1934).
5. Swain, C. G., J. Am. Chem. Soc., 72, 4578 (1950).
6. Swain, C. G., Brown, J. F., J. Am. Chem. Soc., 74, 2534 (1952).
7. Swain, C. G., Brown, J. F., J. Am. Chem. Soc., 74, 2538 (1952).
8. Hudson, C. S., Dale, J. K., J. Am. Chem. Soc., 39, 320 (1917).
9. Bates, F. J., "Polarimetry and Saccharimetry of the Sugars", United States Government Printing Office, Circular C440, 1942, p. 462.
10. Job, P., Ann. Chim. 9, 113, (1937).
11. Bell, R. P., J. Chem. Soc., 362 (1949).
12. Bell, R. P., Waind, G. M., J. Chem. Soc., 1979 (1950).
13. Bell, R. P., Waind, G. M., J. Chem. Soc., 2357 (1951).