SOME ANALYTICAL APPLICATIONS OF THE VARIABLE TIME KINETIC METHOD OF ANALYSIS

By

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SOME ANALYTICAL APPLICATIONS OF THE VARIABLE

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TABLE OF CONTENTS

PART I: KINETIC-CATALYTIC TITRATIONS

PART II: VARIABLE TIME KINETIC DETERMINATION OF NITRILOTRIACETIC ACID

LIST OF TABLES

LIST OF FIGURES

PART I

KINETIC-CATALYTIC TITRATIONS

CHAPTER I

INTRODUCTION

Most kinetic analytical methods of analysis are oriented toward the determination of low concentrations of materials in solution by either the so-called constant time or variable time procedure (1, 2, 3). Both are integral methods in which a finite change in reactant or product is measured. The analytical approach under investigation here can be considered a titrimetric adaptation of the variable time method.

Kinetic-catalytic titrations are based on the addition, at a constant rate, of a reactant, and in which the concentration of the sought for species is determined from the volume, amount, or concentration of titrant added to reach a pre-established signal ratio. In the majority of the cases for a general reaction,

$$R + X \xrightarrow{C} \text{products}$$

where C is a catalyst and R is the monitored species, the concentration of X is adjusted to create pseudo-zero order dependence on this reactant. It also generally applies that the rate is first order with respect to R and initial catalyst concentration. The experimental conditions of the approach being investigated here are such that first or second order dependence in R is retained (even though its order may be any order without affecting the general aspects of the technique), but X is added at a constant rate so as to create a "concentration ramp" as the addition

proceeds. The order of the reaction with respect to X must, however, be other than zero.

Kinetic-catalytic titrations appear to be useful analytical tools for the development of methods to fill existing gaps in concentration ranges. This technique may help move the kinetic approach of analysis to higher concentrations and improve the accuracy and reproducibility at relatively low concentration levels. The approach also appears worthwhile of exploration as a possible analytical probe to study metal chelate catalysis, promotion, and inhibition (4).

To explore the technique and lay the foundation for future work involving kinetic-catalytic titrations, a relatively simple system was needed. It had been reported that ruthenium catalyzes the periodate oxidation of 1,10-phenanthroline ferrous sulfate (5). This is reported as a simple reaction and catalysis appears to be caused by a simple sequence of oxidation-reduction reactions involving periodate, ruthenium, and 1,10-phenanthroline ferrous sulfate. Bromate oxidations were not considered because bromate oxidizes the bromide produced during the reaction and free bromine is formed which participates in the reaction. Moreover, such reactions are often auto-catalytic and also exhibit induction periods. In the selected reaction the periodate is reduced only to iodate (6) and an induction period was not encountered.

CHAPTER II

EXPERIMENTAL APPARATUS

Mechanical

The analytical applications of kinetic-catalytic titrations requires the addition, at a very constant rate, of small volumes of titrant. The small volume of titrant added avoids the need of correcting for dilution of the sample. To this end a constant speed motor driven syringe apparatus was designed and utilized effectively (7).

The apparatus has the following main parts: a 06700-6S Insco multispeed gearmotor (Insco Corporation, Groton, Mass.) with the following speeds-6, 3, 1.2, 0.6, 0.3, and 0.12 rpm, a mounting plate for the motor, relay and switching circuit, lead-screw system (40 turns/inch), a gas tight syringe (Hamilton Company, Inc., Whittier, Calif.), and a metal bench which serves as the supporting rack for the titration vessel.

In the driving position, the lead-screw is advanced downward pushing the plunger of the syringe downward and thus delivering the titrant into the titration vessel. When the plunger reaches a point close to the end of the syringe, an adjustable screw actuates relay 1 by pressing down microswitch 1. See Figure 1. This immediately reverses the direction of the motor and the lead-screw is raised automatically. When the motor reaches the top, microswitch 2 is engaged by another adjustable screw and relay 2 is actuated which stops the motor.

The plunger of the syringe disengages from the plastic piece which

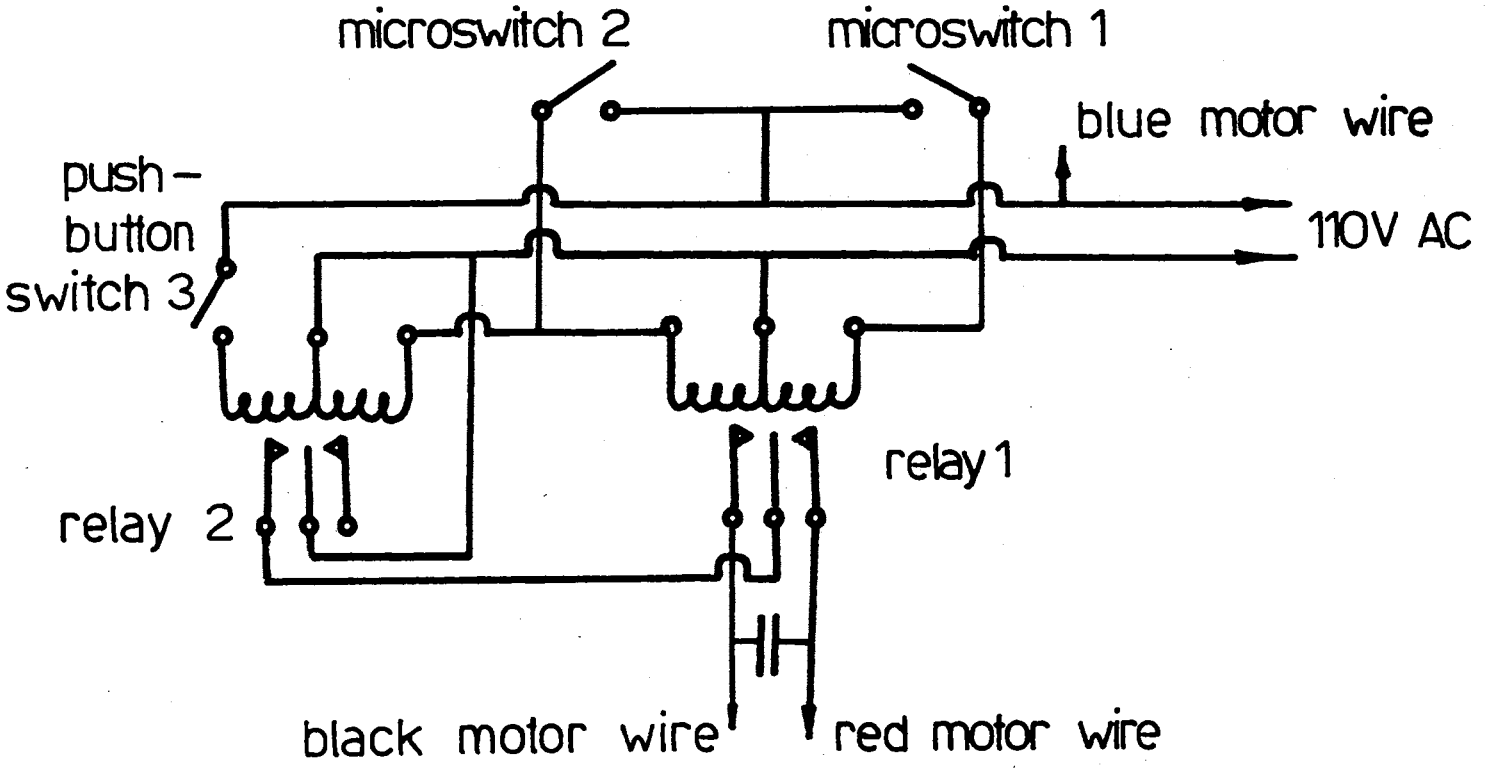

Figure 1. Switching Circuit for "Titration Head"
(From Anal. Biochem., 45, 453 (1972))

was driving it downward as the motor and lead-screw move upward. This allows for easy removal of the syringe for either refilling or cleaning.

When delivery of titrant is wanted again, push-button switch 3 is depressed for approximately five seconds and the delivery is initiated. The depression of switch 3 for five seconds is required to allow the upper adjustable screw to disengage from microswitch 2. After five seconds the motor and lead-screw proceed downward unattended. A side and front view of the apparatus are shown in Figures 2 and 3.

The titrant was introduced into the titration vessel with Teflon (du Pont) tubing (0.22 inch ID) fitted with a Kel-F hub (Hamilton Company, Inc.). This tubing was inserted inside a steel capillary tube (1/16 inch ID) to assure a rigid and therefore reproducible positioning of the tip in the titration vessel.

The precision of the rate of delivery of the apparatus was evaluated by weighing a delivered volume of mercury and relating this to the number of seconds elapsed (7). The amount of mercury delivered was usually one or two drops. The syringe and timer (Precision Time-It, Precision Scientific Co., Chicago, Ill.) were actuated simultaneously. A summary of these statistical results are shown in Table I. Each individual run for a given motor speed represents different length portions of the syringe and lead-screw. The reproducibility for the delivery of the total volume (0.250 or 0.500 ml) was better than 0.2%.

As the samples were monitored spectrophotometrically, the solutions were circulated from the titration vessel through a flow cell (Scientific Cell Company, Forest Hills, N.Y.). A Beckamn DB spectrophotometer (Beckman Instruments, Inc., Fullerton, Calif.) was used to monitor the samples.

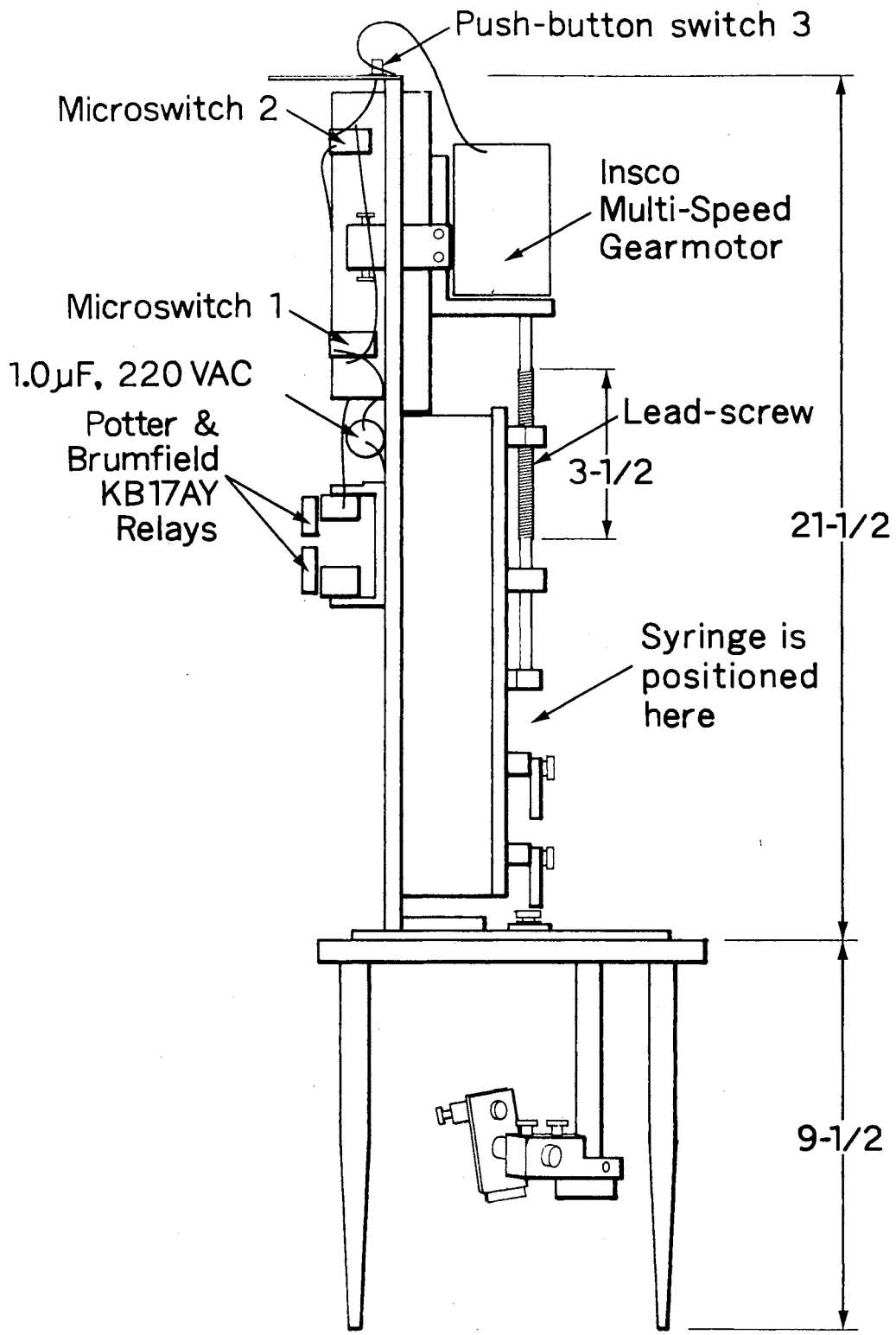

Figure 2. Side View of "Titration Head." Measurements in Inches. (From Anal. Biochem., 45, 453 (1972))

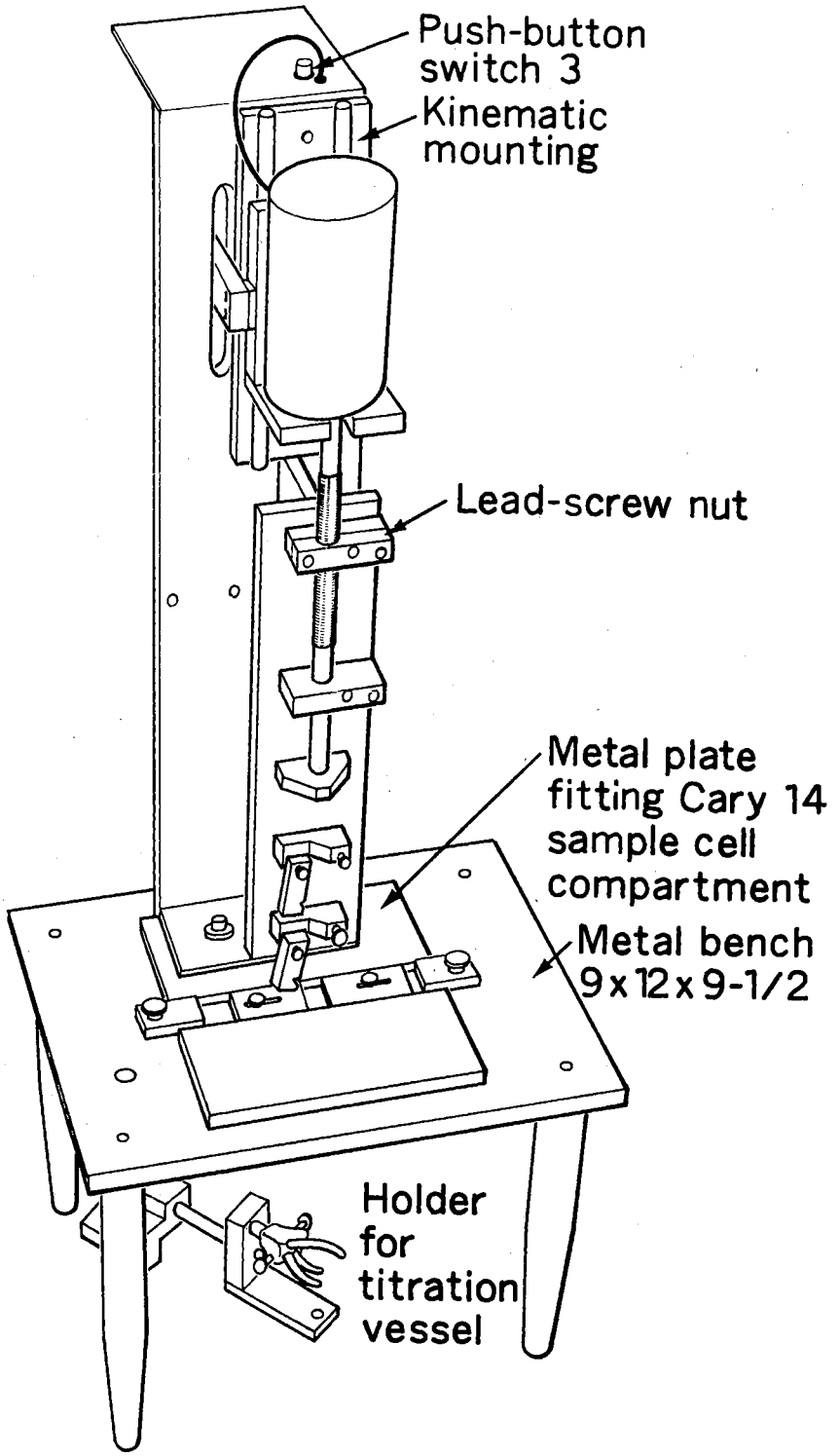

Figure 3. Perspective-Frontal View of "Titration Head". Measurements in Inches. (From Anal. Biochem., 45, 453 (1972))

TABLE I

PERFORMANCE DATA AT 24-26°C FOR DELIVERY OF MERCURY[a] (ALL CALIBRATIONS MADE USING A 0.250 ML SYRINGE, EXCEPT AS NOTED)
(FROM ANAL. BIOCHEM. 45, 453 (1972))

Number of tests (n)	Motor speed, rpm	Mean x 10^4 ml/sec	Max. x 10^4 ml/sec	Min. x 10^4 ml/sec	Standard Dev.[b] x 10^4
30	6	2.670_5	2.712_8	2.617_2	0.0189
30	3	1.329_8	1.417_5	1.310_1	0.0109
16	1.2	0.5290_7	0.5323_9	0.523_9	0.00256
17	0.6	0.2650_9	0.2695_2	0.2625_2	0.00172
15	0.3	0.1324_8	0.1340_3	0.1313_8	0.00112
15[c]	0.3	0.2650_9	0.2695_2	0.2652_2	0.00178
15[c]	0.12	0.1054_1	0.1062_8	0.1045_6	0.00049

[a] Mercury density taken as 13.53_3 gm/ml.

[b] Degrees of freedom: n-1.

[c] 0.500 ml syringe used.

A Variable Speed Masterflex Tubing Pump (Cole-Parmer Instrument and Equipment Co., Chicago, Ill.) was utilized to circulate the solutions through the spectrophotometer. This is a peristalic pump that provides virtually pulseless output and thus a continuous flow of solution from the titration vessel to the spectrophotometer. The solutions were transported by Viton tubing (1/16 inch) and the flow rate of the pump was regulated by a Masterflex SCR Controller (Cole-Parmer, Chicago).

Electronic

As mentioned previously, the concentration of the chemical species under investigation is determined from the volume, amount, or concentration of titrant added to reach a pre-established signal ratio. Since the titrant is added at a constant rate, a measure of the elapsed time between these two pre-established signals would be directly proportional to the volume of titrant added between these two signals. The digital electronic collection of the elapsed time as the system evolved between two pre-established chemical compositions was accomplished by means of a double switch network in conjunction with a digital timer.

The major components of the electronic network are three operational amplifiers, Zener diodes in the feedback loop of two of them (OA2 and OA3), and two reference potential sources. See Figure 4. The operational amplifiers, power supply, reference potential sources, and timer-counter were modular electronic components from McKee-Pedersen Instruments, Danville, Calif. A console (MP-1001) housed the regulated D.C. power supply (MP-1002). The digital timer (MP-1029) recorded the elapsed time with a reliability of ±0.1 seconds. Figure 5 is a pictorial diagram of the switching network connections.

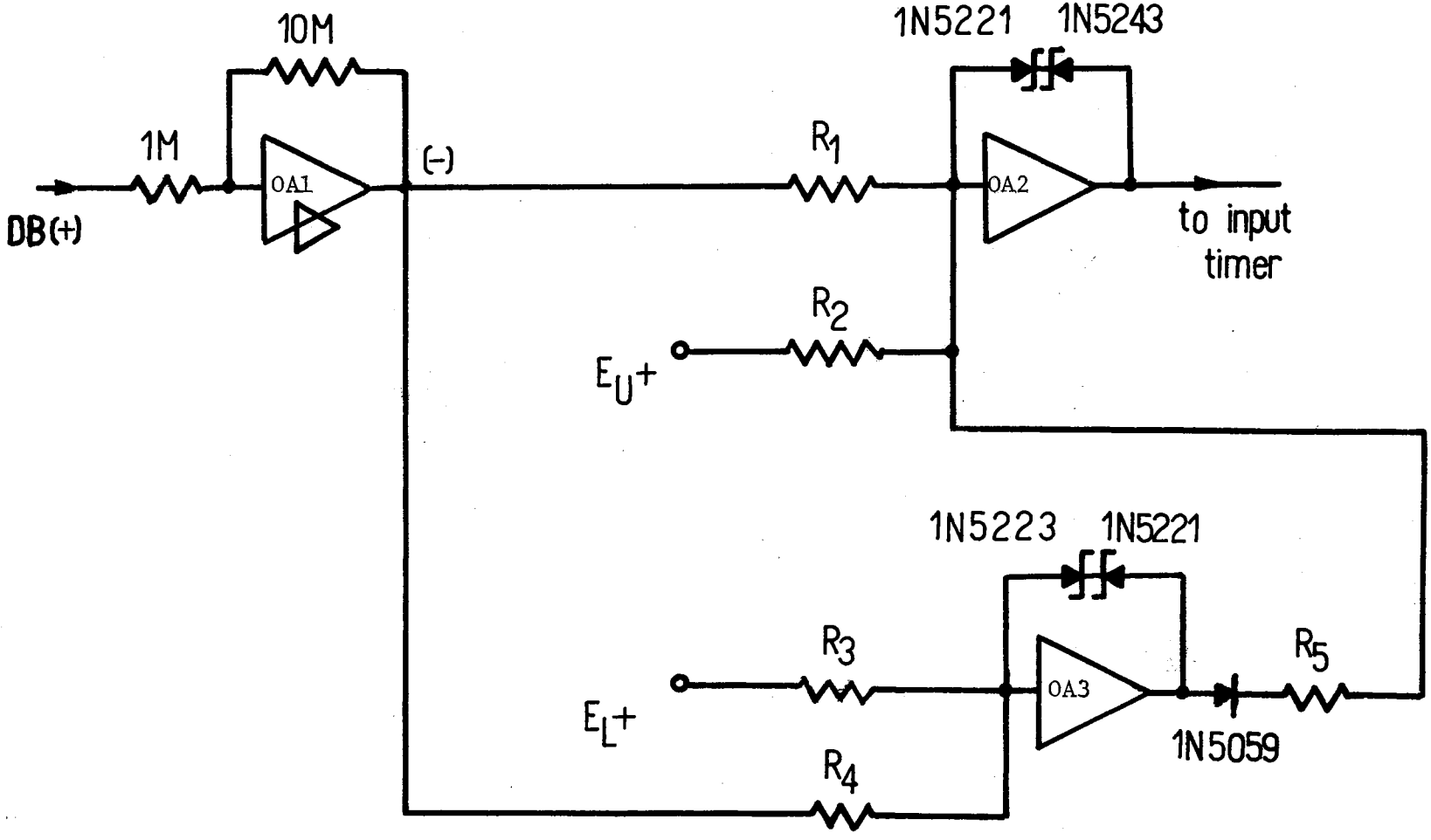

Figure 4. Double Switch Network. E_U and E_L are the reference potential sources from an MP-1008 millivolt source. R_1 to R_5: 10K

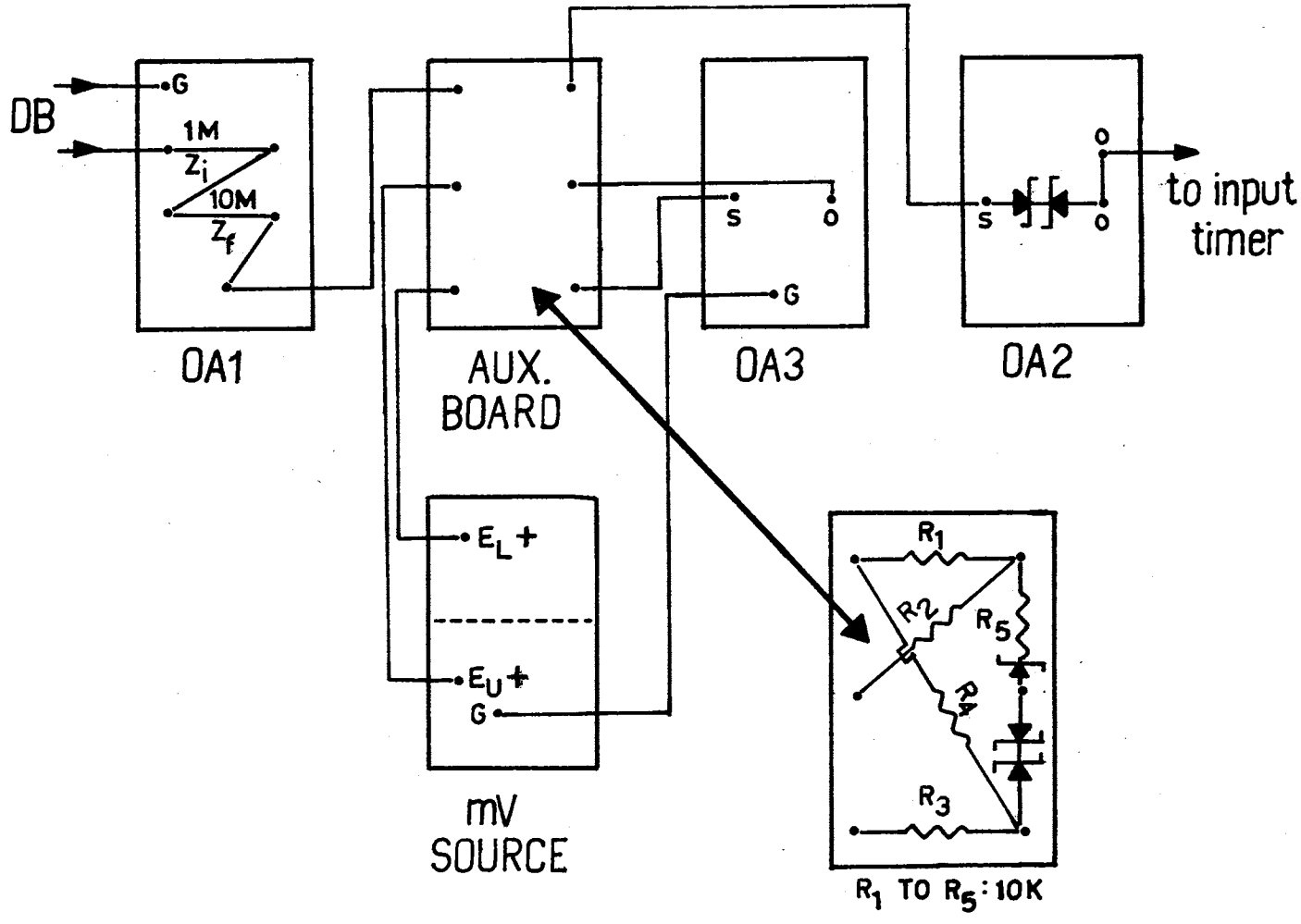

Figure 5. Pictorial Working Diagram of Double Switch Network

OA1 (MP-1031) is a very high gain chopper stabilized D.C. amplifier. OA2 and OA3 (MP-1006A) are regular high gain amplifiers and the maximum output (saturation) potential of all of these amplifiers is ±15 volts (8).

A semiconducting diode is prepared by having an n-type and a p-type junction in a crystal. When the n-type portion is connected to the negative terminal and the p-type is connected to the positive terminal of a voltage source, the diode is said to be forward-biased. Under these conditions a current flows easily due to the low resistance across the n-depletion region-p configuration. A diode is reverse-biased when the terminals are reversed and only a very small amount of current flows because of the high resistance (enlargement of the depletion region). These characteristics of diodes used in conjunction with OA2 and OA3 function as a switch in operating the digital timer.

The negative signal, with respect to ground, coming out of the Beckman DB spectrophotometer ranges from 0 volts to approximately 0.09 volts between 0 and 100% transmittance respectively. If the absorbance reading is equal to 1.0, the output potential of the spectrophotometer is approximately 9 mV. This small signal needs amplification before being fed into the switching network to fall within reasonable range of the reference potential sources. This was achieved by multiplying the spectrophotometer signal by ten with OA1 as shown in Figure 4. Since operational amplifiers have a high input impedance and a low output impedance, OA1 also served to isolate the high impedance of the spectrophotometer.

The maximum output voltage from OA2 and OA3 is ±15 volts as mentioned previously, and any potential greater than ±12 volts, with respect

to ground, applied to the input of the timer-counter keeps it from counting (9).

E_L and E_U are the positive reference potentials from a mV source (MP-1008) and they correspond to the higher absorbance and the lower absorbance values respectively. The values of both reference potentials were periodically controlled and evaluated by means of a Universal Digital Instrument (Heath Company, Benton Harbor, Mich.).

Basically, OA2 is the main switching device in the electronic network. This amplifier either keeps the timer off, actuates it, or stops it from counting depending upon the sign of the input potential.

At the beginning of a determination the absolute value of the signal from the spectrophotometer, E_S, is less than E_L. Referring to Figure 4, the input voltage to OA3 is positive since $|E_L| > |E_S|$. As OA3 inverts the signal of the signal, the output voltage is negative. The feedback impedance in the feedback loop of OA3 is very large compared to the input impedance. Thus the gain of OA3 is large and the amplifier is driven to saturation and the output potential is -15 volts. This signal is added to E_U and E_S before entering OA2. The net output of OA2 until $|E_L| = |E_S|$ is +15 volts and the timer is off. As soon as $|E_S| = |E_L|$ the input voltage from OA3 to OA2 is very small and positive. This makes the output of OA3 small and therefore, the timer starts counting. Diode 1N5059 isolates OA3 when $|E_S| \geq |E_L|$. The timer continues to count while $|E_U| \geq |E_S| \geq |E_L|$. When $|E_S| \geq |E_U|$ OA2 is again driven to saturation by the high impedance in the feedback loop of OA2. This saturation potential is again greater than the 12 volts required to stop the timer and the timer stops counting.

Titration curves were obtained with a Sargent SRL Recorder (log-

arithmic gears) (E. H. Sargent and Company, Chicago, Ill.) connected to the output of OA1. These curves were used to manually compute the elapsed time before the double switch network was completely perfected and to inspect the general shape of the titration curves or signal-time displays.

CHAPTER III

THEORY

For every catalyzed reaction there are two contributions to the total rate of the overall reaction: the uncatalyzed reaction and the catalyzed one which proceed simultaneously. A general reaction may be expressed as:

$$R + X \xrightarrow{C} \text{products}$$

Let X be the titrant, R is the reactant whose concentration is followed with time, and C is a catalyst.

Assuming first order dependence on X, the total reaction rate may be written as follows:

$$-\frac{d[R]}{dt} = k_u[R][X] + k_c[R][X][C]_o \qquad (1)$$

in which:

$$\text{rate of uncatalyzed reaction} = k_u[R][X]$$

and

$$\text{rate of catalyzed reaction} = k_c[R][X][C]_o$$

k_u and k_c are constants for the system and experimental conditions considered and C_o is the original catalyst concentration and the chemical species to be determined.

Equation (1) can be written as:

$$- d[R] = k_u[R][X]dt + k_c[R][X][C]_o\, dt \qquad (2)$$

The instantaneous concentration of X, [X], is given by:

$$[X]_t = C_x - ([R]_o - [R]_t) \qquad (3)$$

where C_x = total concentration of titrant added, $[R]_o$ is the initial dye concentration, and $[R]_t$ is the dye concentration at time t. Since X is added at a constant rate, $dC_x/dt = k$ or $C_x = kt$ where k is the rate of addition of titrant in M/minute. Equation (3) can now be written as:

$$[X]_t = kt - [R]_o + [R]_t \qquad (4)$$

Taking the derivative of both sides and solving for dt, Equation (4) becomes:

$$dt = \frac{d[X] - d[R]}{k} \qquad (5)$$

Substituting Equation (5) into Equation (2) yields:

$$- d[R] = \frac{k_u}{k}[R][X]\,(d[X] - d[R]) + \frac{k_c}{k}[R][X][C]_o\,(d[X] - d[R]) \qquad (6)$$

Expanding and rearranging Equation (6), the following relationship is obtained:

$$- d[R] = d[X]\left(\frac{k_u}{k}[R][X] + \frac{k_c}{k}[R][X][C]_o\right) - d[R]\left(\frac{k_u}{k}[R][X] + \frac{k_c}{k}[R][X][C]_o\right) \qquad (7)$$

Rearranging again and factoring, Equation (7) becomes

$$-d[R]\left[1-\left(\frac{k_u}{k}[R][X]+\frac{k_c}{k}[R][X][C]_o\right)\right]=d[X]\left(\frac{k_u}{k}[R][X]+\frac{k_c}{k}[R][X][C]_o\right) \quad (8)$$

and

$$-\frac{d[R]}{d[X]} = \frac{\frac{k_u}{k}[R][X]+\frac{k_c}{k}[R][X][C]_o}{1-\frac{k_u}{k}[R][X]-\frac{k_c}{k}[R][X][C]_o} \quad (9)$$

or

$$-\frac{d[X]}{d[R]} = \frac{1}{\frac{k_u}{k}[R][X]+\frac{k_c}{k}[R][X][C]_o}-1 \quad (10)$$

Since generally $\frac{1}{\frac{k_u}{k}[R][X]+\frac{k_c}{k}[R][X][C]_o} >> 1$, Equation (10) can be written as:

$$-\frac{d[R]}{d[X]} \approx \frac{k_u[R][X]+k_c[R][X][C]_o}{k} \quad (11)$$

Rearranging Equation (11) gives:

$$-\frac{d[R]}{[R]} \approx \left(\frac{k_u}{k}+\frac{k_c}{k}[C]_o\right)[X]d[X] \quad (12)$$

If the reaction is first order with respect to the concentration of R and Equation (12) is integrated between the two pre-established signals, $[R]_1$ and $[R]_2$, and between $[X]_1$ and $[X]_2$, the following expression is obtained:

$$\ln\frac{[R]_1}{[R]_2} \approx \left(\frac{k_u}{k}+\frac{k_c}{k}[C]_o\right)\left(\frac{[X]_2^2-[X]_1^2}{2}\right) \quad (13)$$

Considering any other order with respect to the concentration of R and again integrating an equation analagous to Equation (12) between $[R]_1$ and $[R]_2$ and $[X]_1$ and $[X]_2$, Equation (12) becomes:

$$\frac{[R]_1^{1-m} - [R]_2^{1-m}}{1-m} \approx \left(\frac{k_u}{k} + \frac{k_c}{k}[C]_o\right)\left(\frac{[X]_2^2 - [X]_1^2}{2}\right) \qquad (14)$$

with $m \geq 2$. Since $[R]_1$ and $[R]_2$ are kept constant from one titration to another, inspection of Equations (13) and (14) shows that the ratios

$$\ln \frac{[R]_1}{[R]_2} \quad \text{and} \quad \frac{[R]_1^{1-m} - [R]_2^{1-m}}{1-m}$$

are both constants. Hence, Equations (13) and (14) can be rewritten as:

$$K \approx \left(\frac{k_u}{k} + \frac{k_c}{k}[C]_o\right)\left(\frac{[X]_2^2 - [X]_1^2}{2}\right) \qquad (15)$$

where K equals either $\ln([R]_1/[R]_2)$ or $([R]_1^{1-m} - [R]_2^{1-m})/(1-m)$. This shows that Equation (15) is valid irregardless of the order ($m \geq 1$) of the reaction with respect to the concentration of R. The only difference is the magnitude of the constant K. In the particular case of the periodate oxidation of Ferroin catalyzed by ruthenium, and as reported by Ottaway et. al. (13), K would correspond to a second order reaction in R and K would be of the magnitude of -2×10^5.

Rearranging Equation (15), the following expression is obtained:

$$\frac{2Kk}{k_u + k_c[C]_o} \approx [X]_2^2 - [X]_1^2 \qquad (16)$$

If $k_c[C]_o/k_u \geq 100$, then $k_u + k_c[C]_o \approx k_c[C]_o$. Initial rate measurements for the particular case of the Ferroin oxidation by periodate ions catalyzed by ruthenium show that $k_c[C]_o/k_u \geq 1000$, satisfying the condition for simplification. For 1:1 stoichiometry*: $[X]_1 = kt_1 - a$ and $[X]_2 = kt_2 - b$ where $([R]_o - [R]_1) = \underline{a}$ and $([R]_o - [R]_2) = \underline{b}$. Equation (16) now becomes:

$$\frac{2Kk}{k_c[C]_o} \approx k^2(t_2^2 - t_1^2) - 2k(t_2b - t_1a) + (b^2 - a^2) \qquad (17)$$

Considering a typical titration curve, t_1 and t_2 are approximately equal to 0.13 and 0.25 minutes respectively and $a = 1 \times 10^{-5}$ $\underline{M}$ and $b = 3 \times 10^{-5}$ $\underline{M}$. Rearranging Equation (17) so that

$$\frac{2Kk}{k_c[C]_o} - k^2(t_2^2 - t_1^2) - (b^2 - a^2) \approx -2k(t_2b - t_1a) \qquad (18)$$

and making the appropriate substitutions, Equation (18) becomes:

$$\frac{2Kk}{k_c[C]_o} \approx -2k(t_2b - t_1a) \qquad (19)$$

since $2Kk/k_c[C]_o >> k^2(t_2^2 - t_1^2)$ and $(b^2 - a^2)$. Substituting $([R]_o - [R]_1)$ and $([R]_o - [R]_2)$ for $\underline{a}$ and $\underline{b}$ respectively and rearranging Equation (19), one obtains:

$$\frac{2Kk}{k_c[C]_o} - 2k([R]_2t_2 - [R]_1t_1) \approx -2k[R]_o(t_2 - t_1) \qquad (20)$$

---

*Except for rather high stoichiometric ratios, the following derivation is also valid for stoichiometric ratios other than 1 with minor corrections for the $[X]_i$ expressions which would not invalidate the approximations used.

As $[R]_1 \approx 9 \times 10^{-5}$ M and $[R]_2 \approx 7 \times 10^{-5}$ M, Equation (20) can be rewritten as:

$$\frac{2Kk}{k_c[C]_o} \approx -2k(t_2 - t_1)[R]_o \qquad (21)$$

since $2Kk/k_c[C]_o >> 2k([R]_2t_2 - [R]_1t_1)$. Solving Equation (21) for $[C]_o$, the following expression is obtained:

$$[C]_o \approx \frac{K}{-[R]_o k_c \Delta t} \qquad (22)$$

where $(t_2 - t_1) = \Delta t$.

Equation (22) shows a linear relationship between the original catalyst concentration, $[C]_o$, and the reciprocal of the elapsed time required for the system to evolve between the two pre-established signals, $[R]_1$ and $[R]_2$. This elapsed time, Δt, can be collected by means of the double switch circuit and electronic timer. A linear relationship should be obtained between $[C]_o$ and $1/\Delta t$ and this should be suitable for preparing working curves for the determination of low concentrations of catalyst in solution.

From Equation (20), it can be seen that a linear relationship would be obtained by plotting $[C]_o$ vs. $([R]_2t_2 - [R]_1t_1)$ since $[R]_1$ and $[R]_2$ are both constants from one titration to another. t_1 and t_2 would have to be measured from a strip chart recorder and the convenience of the double switch circuit to measure Δt would be lost.

First order dependence with respect to $[C]_o$ was assumed in this derivation. Second order dependence on $[C]_o$ would result in a linear relationship between $[C]_o$ and $(1/\Delta t)^{1/2}$.

Justification for calling kinetic-catalytic titrations a titrimetric adaptation of the variable time procedure can be shown in the following manner. A known quantity of titrant, measured as a function of Δt, is added to an undetermined quantity of catalyst. This catalyst concentration is in turn determined from the known amount of titrant added between two pre-established signals.

CHAPTER IV

EXPERIMENTAL PROCEDURE

Reagents

Ruthenium trichloride, hydrate: (Alpha Inorganics, Inc., Beverly, Mass.) Used without further purification. Standardized by reduction to metallic ruthenium (10).

1,10-Phenanthroline ferrous sulfate (Ferroin): (G. Frederick Smith Chemical Company, Columbus, Ohio.) Used without further purification.

Sodium meta periodate: (Fisher Scientific Company, Fair Lawn, N.J.) Certified A.C.S. grade used without further purification. Standardized by titration with thiosulfate (11).

Perchloric acid: (Allied Chemical Company, Morristown, N.J.) Reagent grade (70%) used without further purification.

Procedure

The reaction selected for the preliminary investigation of kinetic-catalytic titrations was the periodate oxidation of 1,10-phenanthroline ferrous sulfate which is catalyzed by small amounts of ruthenium. Ferroin is commonly used to designate 1,10-phenanthroline ferrous sulfate and this name will be used hereafter. In the reduced form Ferroin is blood red and is oxidized to a pale blue color by the periodate. This color change was monitored spectrophotometrically at a wavelength of 505 nm.

Fresh ruthenium solutions were prepared daily from a standardized stock solution. The periodate solutions were prepared in 0.1 N H_2SO_4 to increase their stability (12). Periodically the concentration of the periodate was checked by titrating with thiosulfate (11). The water used throughout the investigation was purified by redistilling de-ionized distilled water through an all-borosilicate glass still equipped with a quartz immersion heater.

A 0.500 ml syringe was used to introduce the periodate into the reaction vessel. Care was taken to ensure that there was no bubbles in the syringe which would interrupt the addition of the titrant at a constant rate.

The reproducibility of the titration curves depended to some extent upon the geometry of the titration vessel. See Figure 6. This configuration allowed for better mixing of the reactants before the solution was monitored in the spectrophotometer. The circulation rate of the reactants, regulated by the Masterflex SCR Controller, was not critical. The only problem arose when the speed was too fast. This caused tiny bubbles to develop which collected on the walls of the flow cell.

All titrations were carried out in 0.01 M $HClO_4$ solution and the Ferroin concentration was 1.0×10^{-4} M. This produced an absorbance value of approximately 1.0. The reagents were put in the titration vessel in the following sequence: (1) six ml of purified water, (2) one ml of $HClO_4$, (3) one ml of ruthenium solution, and (4) two ml of Ferroin. This sequence prohibited the Ferroin from being put into the titration vessel at too low a pH as to produce precipitation of the perchlorate salt of Ferroin.

Since there was some slight drift in the spectrophotometer, it was

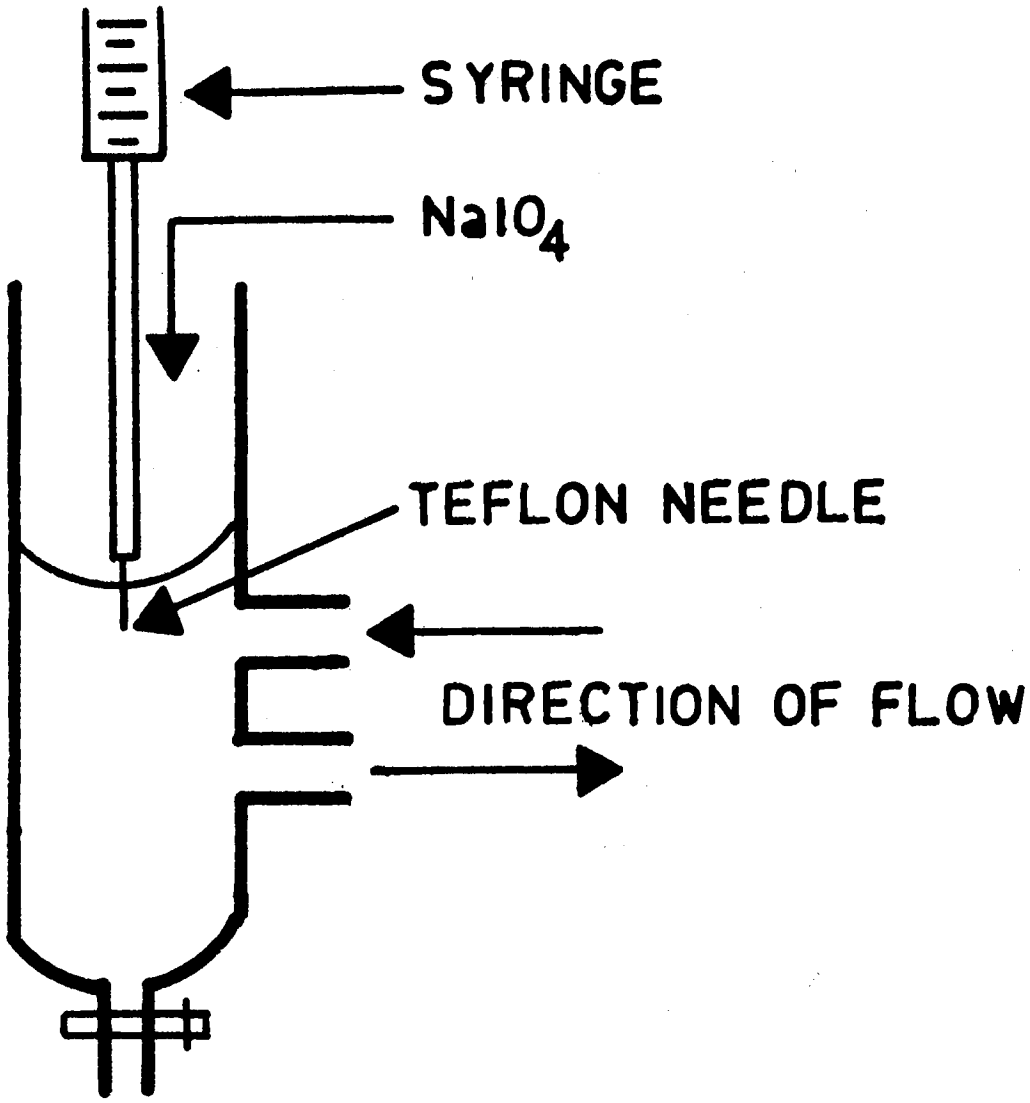

Figure 6. Titration Vessel

zeroed before titration by placing purified water in the reference cell and circulating purified water through the sample cell. The initial absorbances were thus reproducible and the reference potentials were constant from one titration to another. The entire system was rinsed three or four times with purified water after each titration to remove the residual solution.

CHAPTER V

RESULTS AND DISCUSSION

The ruthenium catalyzed periodate oxidation of Ferroin can be formulated as follows:

$$2[Fe(1,10\text{-phen.})_3]^{+2} + \underset{(H_5IO_6)}{H_4IO_6^-} + 2H^+ \xrightarrow{Ru} 2[Fe(1,10\text{-phen.})_3]^{+3} + IO_3^- + 3H_2O$$

As stated previously, Ferroin forms a precipitate if introduced into the solution at too low a pH. The catalytic method for the determination of ruthenium described by Ottaway et. al. (13) suggests a $HClO_4$ concentration of 0.1 M. This acid concentration was tried and a precipitate developed which was probably the [Ferroin^{+2}, $2ClO_4^-$] complex. Various lower concentrations of $HClO_4$ were investigated and satisfactory results were obtained by running the titrations in 0.01 M $HClO_4$.

Figure 7 shows some typical titration curves obtained from a strip chart recorder. A 2.5% periodate solution was used as the titrant and the rate of delivery was 5.34×10^{-4} ml/second. The rate of the uncatalyzed reaction is almost negligible compared with the catalyzed reaction and the latter reaction is extremely sensitive to changes in concentration of the catalyst. If an induction period is observed, it could be tolerated if the time involved does not create an excess of periodate in the solution. The induction period should also be reproducible so that the amount of periodate added will be the same for replicate titrations.

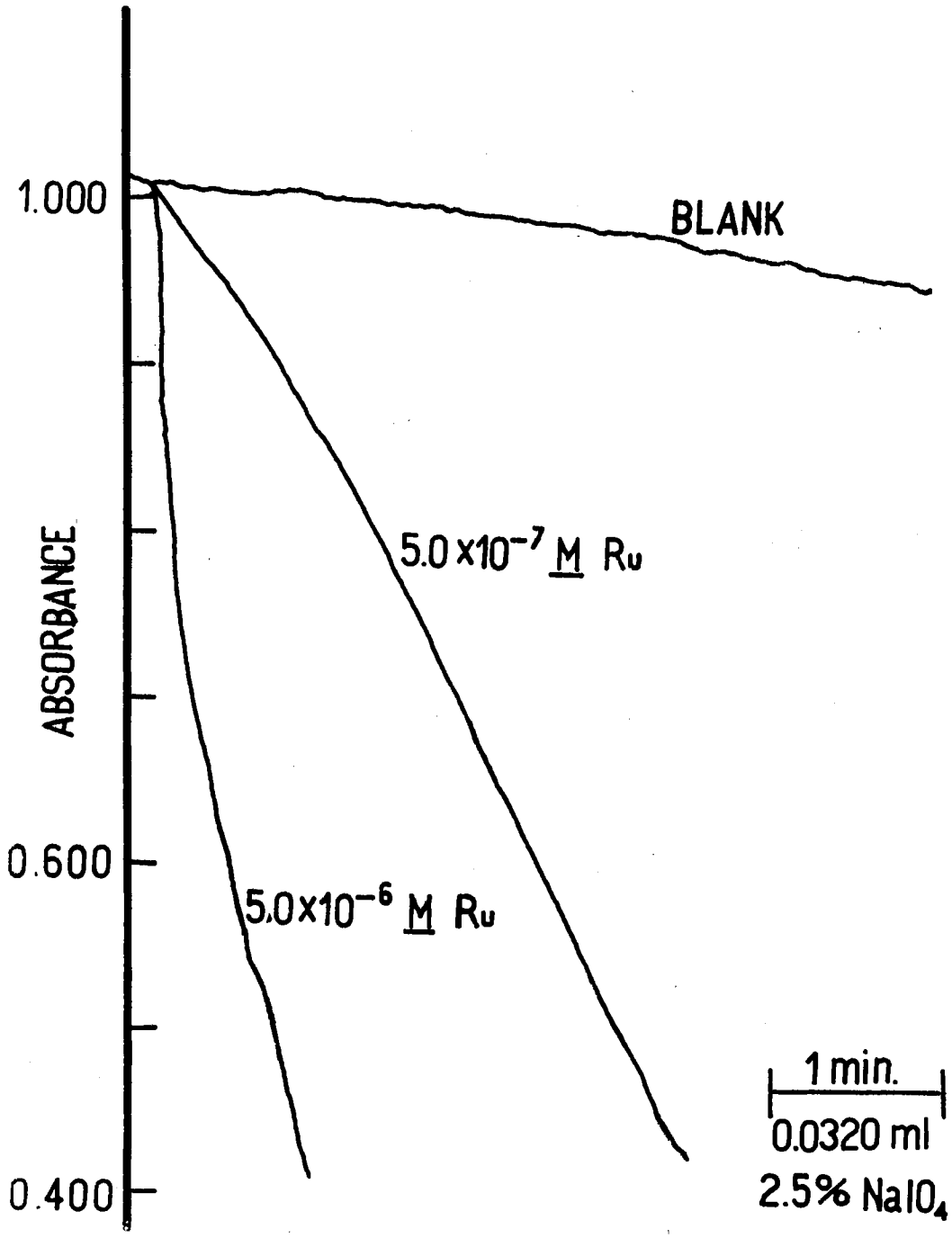

Figure 7. Recorded Titration Curves for the Ferroin-Periodate Reaction Catalyzed by Ruthenium

Before the double switch circuit was utilized for the digital collection of the elapsed time between the two absorbance values, the titration curves were recorded on a strip chart recorder and the elapsed times were manually computed. The output signal from OA1 was sent to the recorder and the addition of titrant and the recorder were actuated simultaneously. Figure 8 shows a working curve obtained by this procedure. Elapsed times were measured between absorbance values of 0.800 and 0.400. A plot obtained from elapsed times collected by means of the double switch circuit, with E_U = 0.2699 volts and E_L = 0.1410 volts (0.510 and 0.780 absorbance readings respectively), is illustrated in Figure 9. It is apparent that the use of the double switch circuit allowed greater precision and also permitted a larger concentration range for the determination of ruthenium.

As this procedure depends upon the reaction being other than zero order with respect to the periodate concentration, the absorbance values were selected near the beginning of the titrations. In this manner the periodate concentration was kept to a minimum, and at the same time the elapsed times could still be quantitatively collected.

Ruthenium solutions ranging in concentrations from 1.0×10^{-6} M to 9.0×10^{-6} M were titrated with 2.5% periodate. Potential values were set at E_U = 0.1795 volts and E_L = 0.1064 volts which correspond to the pre-established absorbance readings of 0.710 and 0.920 respectively. See Figure 10. Between ruthenium concentrations from 5.0×10^{-6} M to 9.0×10^{-6} M, Δt varied from 9.7 seconds to 5.7 seconds. Manually measuring these times would be virtually impossible and the errors involved would be tremendous without the aid of the electronic timer.

At the point where the timer shuts off, approximately 5.6×10^{-4} M

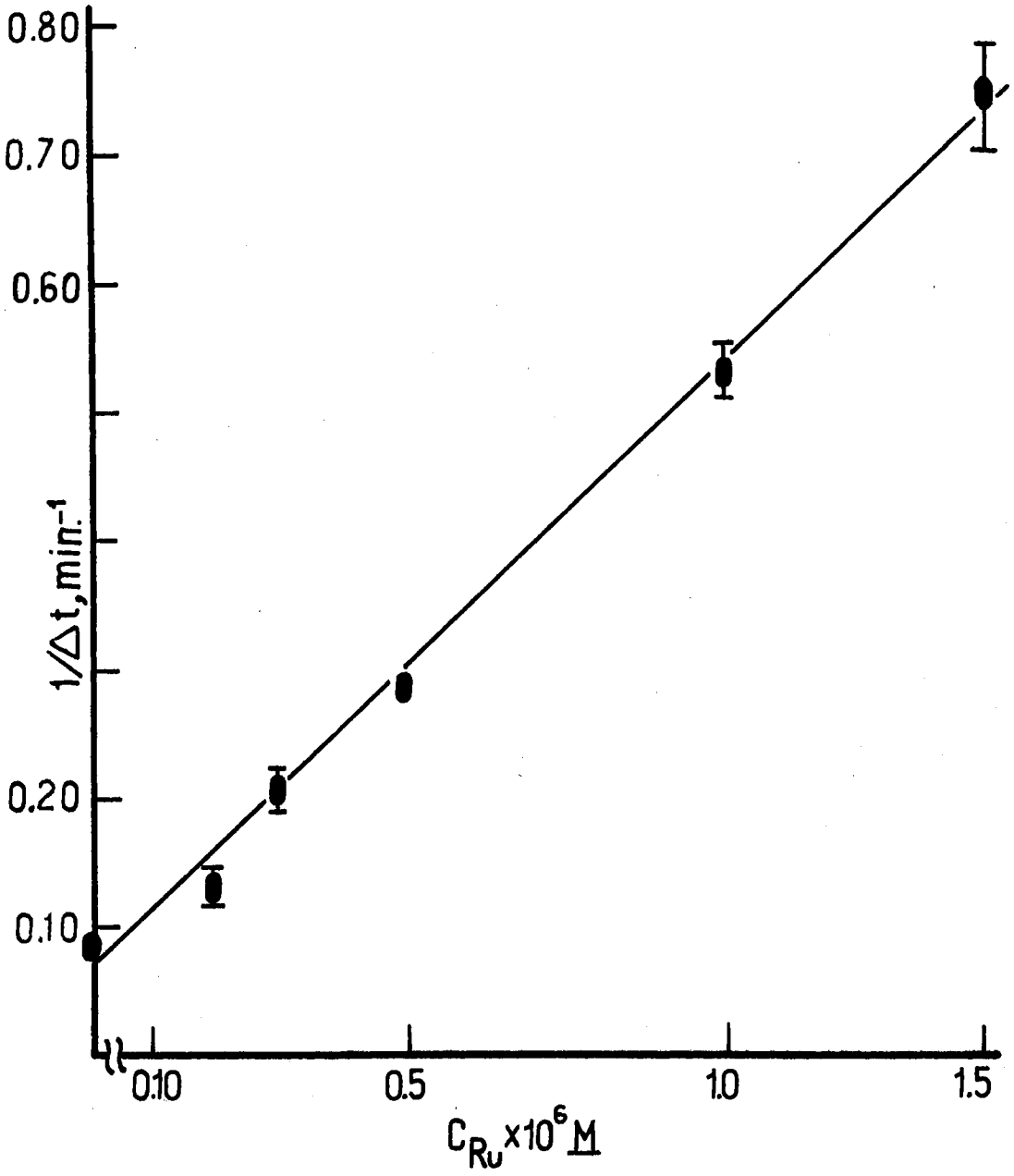

Figure 8. Working Curve for Ruthenium Determination. Elapsed times estimated from recorded titration curves

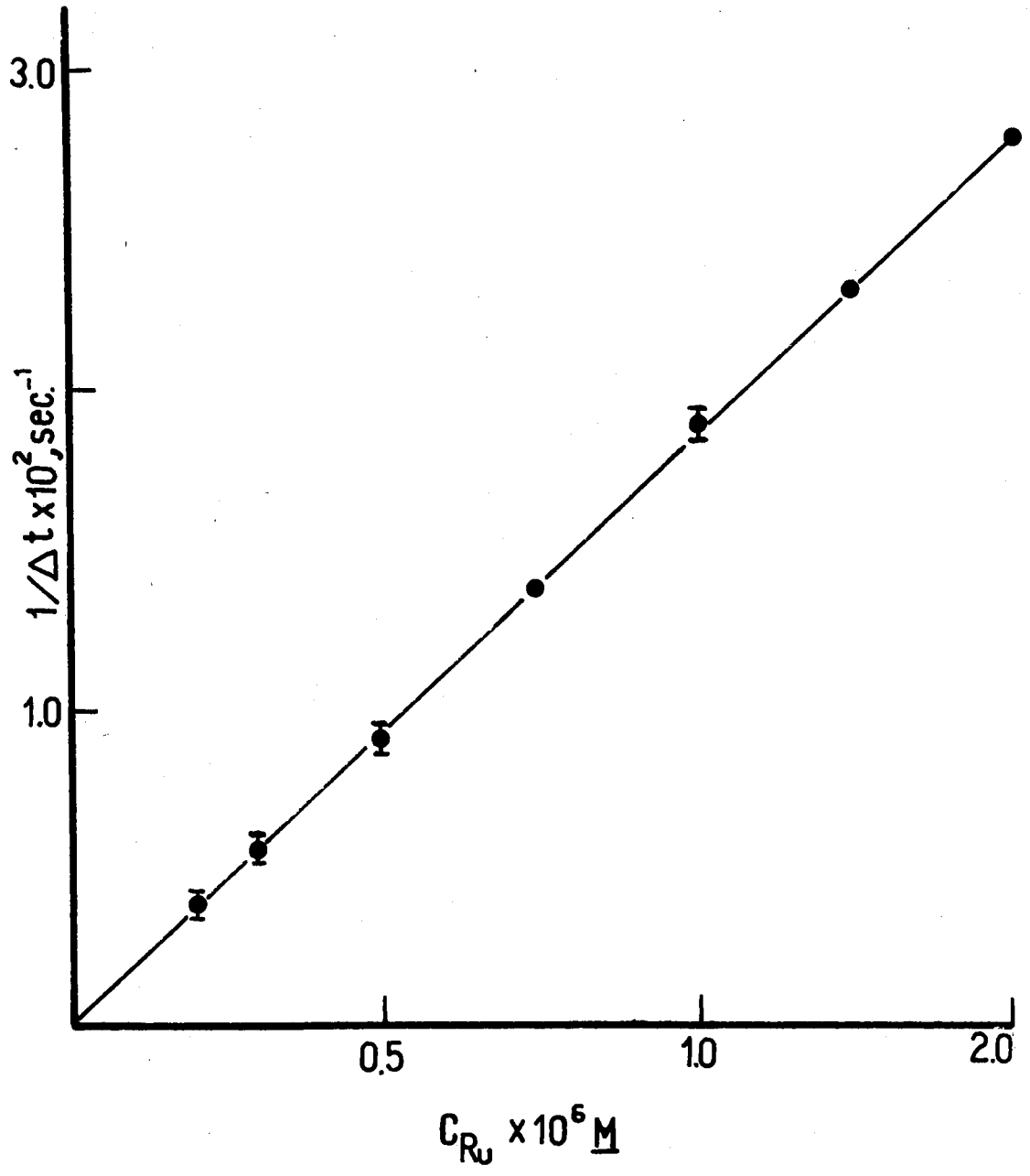

Figure 9. Working Curve for Ruthenium Determination. Elapsed times collected with double switch network and digital timer

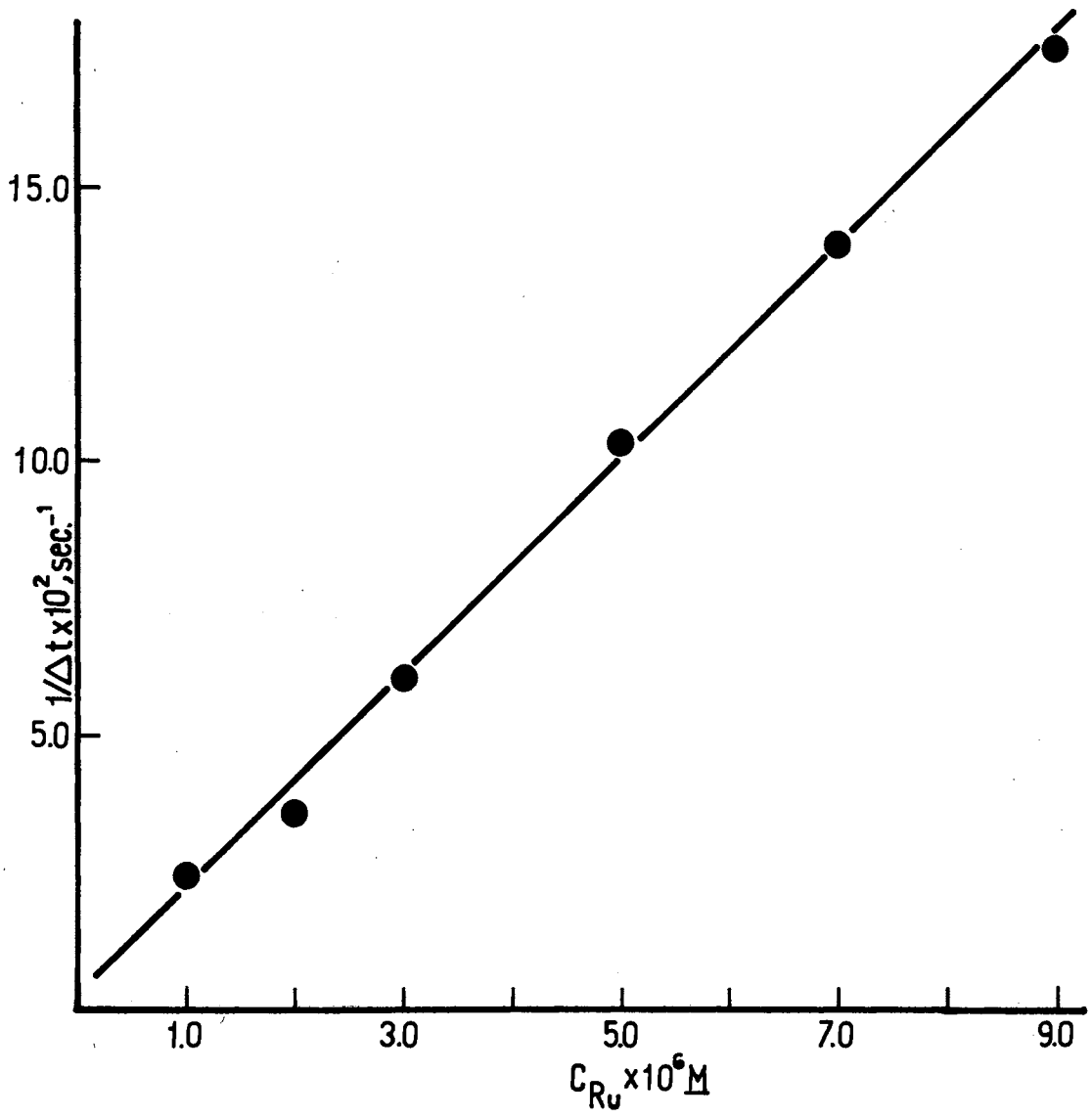

Figure 10. Working Curve for Ruthenium Determination According to Procedure in Text

total periodate had been added to the solution containing the least amount of ruthenium (1.0×10^{-6} M). This is approximately an 8:1 ratio of periodate to Ferroin when the timer shuts off. Since Ferroin and periodate react in a 2:1 ratio, the correction for the amount of periodate consumed was negligible and therefore neglected. All other ruthenium concentrations gave much faster reactions and the total periodate added ranged from approximately 2.4×10^{-4} M to 0.93×10^{-4} M. The ratios of Ferroin concentration to periodate concentration seem to indicate that the reaction cannot be zero order with respect to the periodate concentration under these experimental conditions.

Figure 11 shows a titration curve obtained by stopping the addition of periodate as soon as the reaction started and then re-starting the addition at a later time. If the periodate was in excess so as to create pseudo-zero order dependence, the absorbance should have continued to decrease when the syringe was shut off. This was not the case; furthermore, the oxidation of the Ferroin commenced as soon as periodate was again added.

Since the ratios of Ferroin concentration to periodate concentration when the timer shuts off are quite below the needed excess of periodate for pseudo-zero order dependence in this reagent and considering the previous behavior, assurance of non-zero dependence with respect to the periodate seems to be indicated.

Approximately a 30 second induction period was encountered when titrating the 1.0×10^{-6} M ruthenium solution indicating the catalytic activity of ruthenium may be governed by the ratio of Ru(III) to Ru(IV) in solution. At the beginning of the titration, Ru(III) predominates exclusively and since the ruthenium concentration is low, more periodate

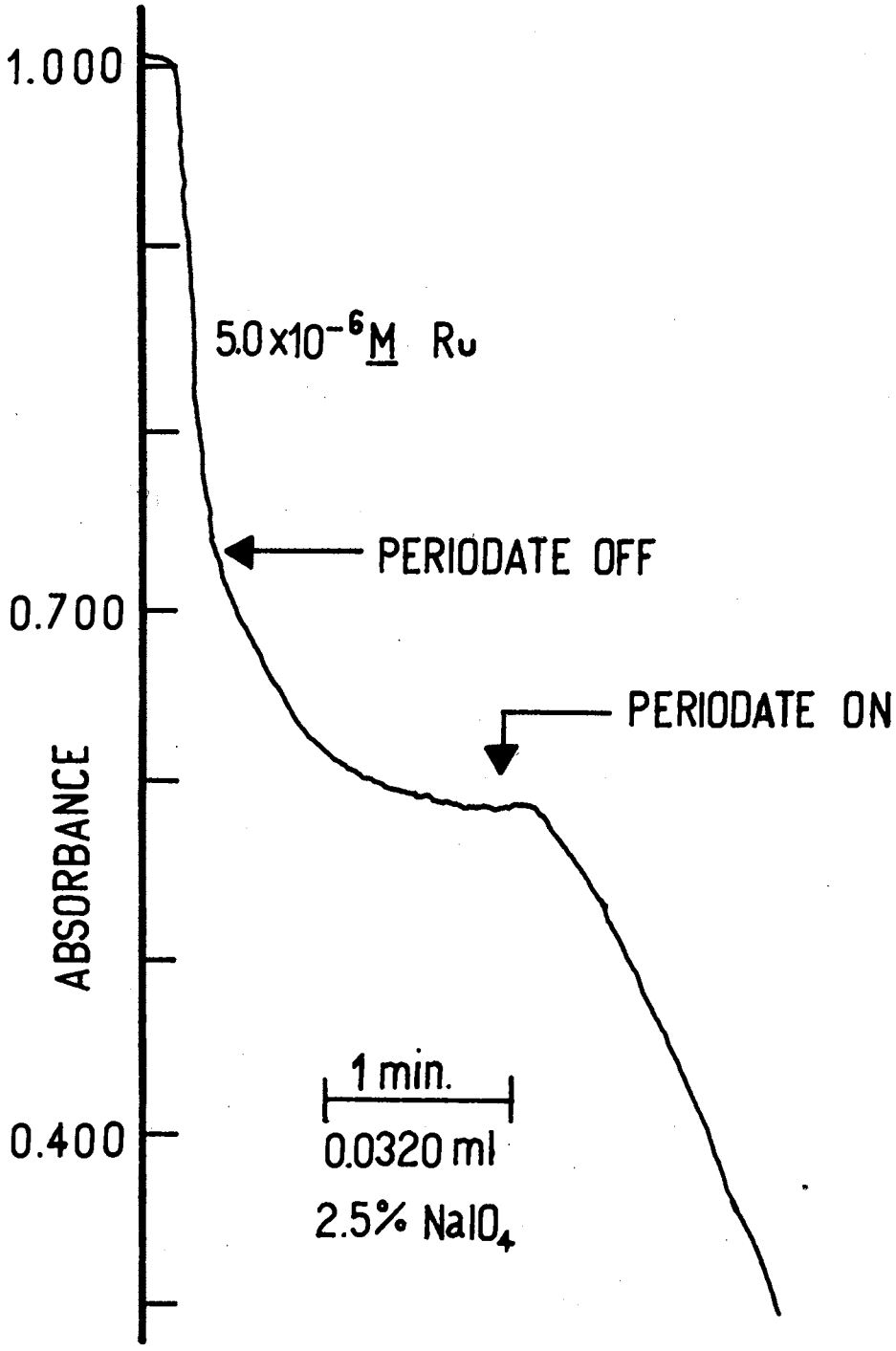

Figure 11. Recorded Titration Curve Showing the Effect of Interrupting the Periodate Addition

is required to reach the proper Ru(III) to Ru(IV) ratio.

The oxidation of ruthenium to the higher oxidation state by periodate was considered to be first order with respect to both the catalyst and the periodate concentrations (14). According to Ottaway et. al. (13) the reaction is first order with respect to the catalyst concentration and second order with respect to the Ferroin concentration. Treatment of the experimental data under these conditions showed that a linear relationship was obtained between the original catalyst concentration, $[C]_o$, and $1/\Delta t$.

The kinetics of this reaction have been studied by Ottaway et. al. as stated in reference (13). The promised detailed publication of their studies, however, has not appeared in the literature as of yet.

Kalinina (14) suggests that the oxidizing species of periodate at pH ≈ 2 for Ru(III) going to Ru(IV) is $H_6IO_6^+$. Haladjian et. al. (15), Turney (16), and Pavlova et. al. (17) do not mention this species irregardless of the pH of the solution and it seems hard to visualize the existance of this species in aqueous solutions of pH 1 to 2. At pH ≈ 2 there is approximately 80% $H_4IO_6^-$ and 20% H_5IO_6 (17).

Since the kinetic-catalytic titration approach heavily depends on assurance of both reactant concentrations affecting the rate, further work on the exploration of this technique depends on the possibility of utilizing rather large concentrations (high absorbance values) of the monitored species. The use of the Cary 14 spectrophotometer with electronically compatible timing networks or specially designed photometric units are instrumental approaches which should be explored as continuation of this work.

PART II

VARIABLE TIME KINETIC DETERMINATION OF NITRILOTRIACETIC ACID

CHAPTER I

INTRODUCTION

Polyphosphates contribute to the eutrophication of streams and lakes by acting as a nutrient for algae, and the largest contributors of polyphosphates to our environment are household detergents. The trisodium salt of nitrilotriacetic acid (NTA) has been used as a partial substitute for polyphosphates in some detergent formulations. NTA was used as a substitute because it was more economical than other chelating agents (18). Also its chelating power is greater than most other complexing agents that could have been utilized (19).

In December of 1970, however, the sale of detergents in the United States containing NTA was halted because of its possible toxicity and carcinogenic effects. The cursory reports on NTA toxicity are, however, confusing and point to the need for future research on the analytical chemistry of NTA. The use of NTA, for instance, has recently been approved by the National Swedish Health and Welfare Board, but they emphasize that more detailed research is needed in several areas where the present evidence of the effects of NTA is ambiguous.

Biochemical and biological studies are being undertaken to evaluate the effects of NTA on the environment (43). All of these considerations point to the potential usefulness of sensitive tests for this aminopolycarboxylic acid. A rapid and efficient method for the determination of low concentrations of NTA would be most advantageous to help further the

research needed to more fully understand the toxicological problems associated with NTA.

Analytical methods for the determination of NTA were first developed for samples of ethylenediaminetetraacetic acid (EDTA) containing low concentrations of NTA. EDTA contaminated with low concentrations of NTA does not give a sharp color change in some complexometric titrations (20).

With infrared analysis a minimum of 1% NTA in 99% EDTA can be detected (21). Daniel et. al. (22) have shown that a polarographic method can be employed to determine NTA at a concentration range from 1.0×10^{-5} M to 1.0×10^{-3} M. Another common impurity in EDTA, along with NTA, is N,N'-ethylenediglycine (EDDA). NTA and EDDA can both be determined in EDTA by a modified polarographic procedure similar to the one previously mentioned (23). This method is feasible if the concentration level of NTA is between 5.0×10^{-5} M and 1.0×10^{-3} M. Farrow and Hill (24) have devised another polarographic technique that is capable of detecting 20 ppm NTA in EDTA.

The potentiometric titration with Zn(II) ions of EDTA-NTA mixtures has been reported and NTA in the 10-100 mg range can be determined (25). Titration of EDTA-NTA mixtures with Fe(III) ions allows the potentiometric determination of 1-2% NTA in EDTA (26).

Doran (27) has reported the determination of NTA and related compounds by rapid paper electrophoreses. Complexometric titrations using Cu(II) or Fe(III) ions as the titrant have been effectively utilized to quantitatively determine NTA above 1.0×10^{-4} M (28,29). Holzapfel et. al. (30) have shown that 10-100 mg of NTA can be determined by a titrimetric procedure involving the oxidation of NTA in the presence of

Fe(III) ions by Ce(IV).

Complexometric titrations employing Cu(II) ions as the titrant have been shown to be effective in detecting NTA in environmental samples (31) and in granular detergents (32) that range in concentration levels from 5000-6000 ppm respectively. As little as 5.0×10^{-5} M NTA in detergent formulations has been reported by Rechnitz et. al. (33) as the lower limit of detection for NTA by using a solid membrane Cu(II) ion selective electrode. Another complexometric method employs the thin layer chromatographic identification and subsequent titrimetric determination of NTA in detergents again using Cu(II) ions as the titrant (34).

Swisher et. al. (35) have reported that NTA in activated sludge effluents can be determined in the concentration range of 5-50 mg/1. A zinc-zincon analytical method for the determination of sequestrants in water and sewage has been developed that is capable of detecting 0.15 ppm NTA (36).

Spectrophotometric analysis of uncomplexed Cu(II) ions has been employed on waste water samples containing 5-12 mg/1 NTA (37). An adaptation of this method allowed the workable concentration of NTA to be reduced to 0.2-3 ppm (38).

Polarographic techniques (39,40,41) have been reported that will determine 0.026-10 ppm NTA in environmental samples. Chau et. al. (42) and Rudline (43) have used gas chromatographic methods for the analysis of lake waters and report detecting NTA as low as 0.01 ppm after concentrating the samples.

Margerum et. al. (44) have reported a kinetic method of analysis that allowed the simultaneous determination of mixtures of aminopolycarboxylic acids from 1.0×10^{-7} M to 1.0×10^{-4} M. The details of this

procedure do not seem to have appeared in the literature.

Recent additions to the literature for the determination of NTA in detergents, natural waters, and sewage samples include polarography (45), gas chromatography (46), high speed ion exchange chromatography (47), a modified zinc-zincon method (48), and a colorimetric procedure (49). All of the above methods, except the colorimetric approach, seem to be capable of detecting NTA as low as 0.05-1 ppm. The colorimetric method was used on detergents and 28% NTA in detergents was quantitatively determined.

Most analytical methods reported for the determination of NTA show disadvantages which can be identified in lack of sensitivity, poor detectability, time requirement for analysis, and pre-treatment of the sample prior to the actual analysis.

This investigation was undertaken to evaluate the determination of low concentrations of NTA by a relatively rapid, simple, and selective procedure. The procedure employed for the determination of small amounts of NTA was the variable time kinetic method of analysis and this procedure should be adaptable to bio-environmental samples quite readily.

CHAPTER II

THEORY

In many homogeneous reactions involving a substance which acts as a catalyst, the concentration of the catalyst is directly (or nearly directly) proportional to the rate of the chemical reaction. The catalytic species can, therefore, be determined from the rate of these reactions.

The variable time procedure is one of the methods employed to estimate the concentration of the catalyst. This procedure is well defined for true metal catalysis (1,2). A general reaction may again be expressed as:

$$R + X \xrightarrow{C} \text{products}$$

where R is the reactant whose concentration is followed with time, X is also a reactant, but it is added in excess to create pseudo-zero order dependence on X, and C is a catalyst. It generally applies that the rate of the reaction is first order with respect to both the concentration of R and the concentration of the catalyst.

The total reaction rate may be expressed as:

$$-\frac{d[R]}{dt} = k_u[R] + k_c[R][C]_o \qquad (1)$$

Rearranging and integrating between $[R]_1$ and $[R]_2$ and t_1 and t_2, Equation (1) becomes:

$$\ln \frac{[R]_1}{[R]_2} = (t_2 - t_1)(k_u + k_c[C]_o \quad (2)$$

If $\ln([R]_1/[R]_2)$ = constant = K and $(t_2 - t_1) = \Delta t$, then:

$$[C]_o = \frac{K}{\Delta t k_c} - \frac{k_u}{k_c} \quad (3)$$

Hence, for true metal catalysis, the original catalyst concentration can be determined from the linear relationship between $[C]_o$ and $1/\Delta t$. This is the variable time integral kinetic method of determination.

Upon addition of a complexing (chelating) agent, the rate of a chemical reaction may be modified further. Three possibilities seem to predominate from a mechanistic viewpoint (4). First, the ligand may subtract the metal from the catalytic cycle and in doing so inhibiting the catalytic path (ligand inhibition). Also, by metal complexation, the rate of a chemical reaction can be accelerated either by true metal complex catalysis or by ligand promotion.

From an equilibrium viewpoint, inhibition seems to be favored when the transition metal complex, in a lower oxidation state, predominantly complexes with the ligand. The catalytic species is thus withdrawn from active participation in the catalytic cycle. Promotion and/or true metal catalysis may be observed when the metal catalyst, in a higher oxidation state, predominantly combines with the complexing agent and enters into the promotion path and/or the catalytic cycle. Ligand promotion is characterized by the ligand being destroyed as the promotion path proceeds.

If the catalyst concentration, C_o, is less than the ligand concen-

tration, C_L, both true metal chelate catalysis and ligand promotion may occur. The total reaction rate for these conditions may be written as:

$$-\frac{d[R]}{dt} = k_u[R] + k_c'[R][C^*L] \tag{4}$$

where C*L is the metal complex and the transition metal catalyst is in a higher oxidation state. Again rearranging Equation (4) and integrating between $[R]_1$ and $[R]_2$ and t_1 and t_2, the following expression is obtained:

$$[C^*L] \propto C_L \approx \frac{K}{\Delta t k_c'} - \frac{k_u}{k_c} \tag{5}$$

Since $[C]_o$ is less than C_L, the concentration of the metal chelate, C*L, is proportional to C_L. Therefore, the original ligand concentration, C_L, can be determined in this manner.

The other possibility, and the one concerned with in this investigation, is when the catalyst concentration is greater than the ligand concentration. In this case, the rate modifying effect of the ligand seems to be mainly due to true metal chelate catalysis. Considering a general reaction:

$$R + X \xrightarrow{C,C^*L} \text{products}$$

the total reaction rate may be written as:

$$-\frac{d[R]}{dt} = k_u[R] + k_c([C]_o - [C^*L])[R] + k_c'[C^*L][R] \tag{6}$$

in which:

rate of uncatalyzed reaction = $k_u[R]$

rate of catalyzed reaction = $k_c([C]_o - [C*L])[R]$

and rate of metal chelate catalyzed reaction = $k_c'[C*L][R]$

k_u, k_c, and k_c' are constants for the system and experimental conditions considered, R is the reactant whose concentration is followed with time, X is a reactant added in excess to assure pseudo-zero dependence, $[C]_o$ is the original catalyst concentration, and C*L is the metal complex providing the rate modifying path.

Equation (6) can be rearranged to:

$$-\frac{d[R]}{[R]} = k_u dt + k_c([C]_o - [C*L])dt + k_c'[C*L]dt \qquad (7)$$

If the concentration of R is followed spectrophotometrically:

$$A = \text{absorbance} = \varepsilon b[R]$$

where ε is the molar absorptivity of R and b is the cell path length. Equation (7) can now be written as:

$$-\frac{dA}{A} = k_u dt + k_c([C]_o - [C*L])dt + k_c'[C*L]dt \qquad (8)$$

Integration of Equation (8) between two absorbance values, A_1 and A_2, and between t_1 and t_2, the following expression is obtained:

$$\ln\frac{A_1}{A_2} = k_u t_2 - k_u t_1 + k_c([C]_o - [C*L])t_2 - k_c([C]_o - [C*L])t_1 + k_c'[C*L]t_2 - k_c'[C*L]t_1 \qquad (9)$$

Since A_1 and A_2 remain constant from one determination to another, $\ln(A_1/A_2)$ = constant = K. Hence, Equation (9) can be written as:

$$K = (t_2 - t_1)\{k_u + k_c([C]_o - [C^*L]) + k_c'[C^*L]\} \quad (10)$$

Equation (10) is independent of the order of the reaction with respect to the concentration of R. This is one of the advantages of the variable time procedure which has been indicated as superior to the fixed time approach (1) in case of a non-linear response of the reaction monitored signal (3).

Solving Equation (10) for $[C^*L]$, the following relationship is obtained:

$$[C^*L] = \frac{K}{(t_2 - t_1)(k_c' - k_c)} - \frac{k_u + k_c[C]_o}{(k_c' - k_c)} \quad (11)$$

Since $[C]_o$ is constant from one determination to another, Equation (11) becomes:

$$[C^*L] = \frac{\text{constant}}{(t_2 - t_1)} - \text{constant}' \quad (12)$$

where $K/(k_c' - k_c) = \text{constant}$ and $(k_u + k_c[C]_o)/(k_c' - k_c) = \text{constant}'$. Equation (12) may be expressed as:

$$[C^*L] \propto 1/\Delta t \quad (13)$$

where $(t_2 - t_1) = \Delta t$. As previously stated, $[C]_o$ is greater than C_L and therefore:

$$[C^*L] \propto C_L \propto 1/\Delta t \quad (14)$$

or

$$C_L \propto 1/\Delta t \quad (15)$$

Relationship (15) shows a linear dependence between C_L and $1/\Delta t$ and this method should be applicable to determine low concentrations of complexing agents in solution.

CHAPTER III

EXPERIMENTAL PROCEDURE

Reagents

Malachite green oxalate: (Eastman Organic Chemicals, Rochester, N.Y.) White label used without further purification.

Manganous sulfate, monohydrate: (J. T. Baker Chemical Company, Phillipsburg, N.J.) Reagent grade used without further purification.

Nitrilotriacetic acid: (Eastman Organic Chemicals, Rochester, N.Y.) White label purified by back precipitation as free acid from basic solution with concentrated $HClO_4$. Standardized by complexometric titration with Cu(II) (50).

Sodium meta periodate: (Fisher Scientific Company, Fair Lawn, N.J.) Standardized by titration with thiosulfate (11).

Phosphate-acetate buffer (pH ≈ 3.5): 35 grams of NaH_2PO_4 and 15 ml of glacial acetic acid diluted to 500 ml with purified water and the pH adjusted with 1.0 M NaOH.

Ethylendiaminetetraacetic acid: (J. T. Baker Chemical Company, Phillipsburg, N.J.) Reagent grade used without further purification.

Interferences

Iminodiacetic acid: (Hampshire Chemical Division, Nashua, N.H.) Purified by back precipitation as free acid from basic solution with concentrated $HClO_4$.

Glycolic acid (purified): (Fisher Scientific Company, Fair Lawn, N.J.)

Glycine: (Matheson Coleman and Bell, Norwood, Ohio.) 99.5% used without further purification.

Sodium tripolyphosphate: (Alpha Inorganics, Inc., Beverly, Mass.) Used without further purification.

Dodecylbenzene sodium sulfonate: (K and K Laboratories, Inc., Plainview, N.Y.) Purified by extraction of saturated solution with isopropyl alcohol and evaporation of oily layer to dryness.

Sodium lauryl sulfate: (K and K Laboratories, Inc., Plainview, N.Y.) USP grade purified by extraction of saturated solution with isopropyl alcohol and evaporation of oily layer to dryness.

Ferric perchlorate: (G. Frederick Smith Chemical Company, Columbus, Ohio.) Reagent grade used without further purification. Standardized by complexometric titration with ethylenediaminetetraacetic acid (50).

Calcium perchlorate hexahydrate: (G. Frederick Smith Chemical Company, Columbus, Ohio) Reagent grade used without further purification. Standardized by complexometric titration with ethylenediaminetetraacetic acid at pH 12 with Eriochrome Blue SE as indicator.

Ortho cresol: (Matheson Coleman and Bell, Norwood, Ohio.) 99+% used without further purification.

Meta cresol: (Fisher Scientific Company, Fair Lawn, N.J.) 95-98% used without purification.

Chlorine: (The Matheson Company, Inc., East Rutherford, N.J.)

Procedure

The mechanical and electronic apparatus, experimental conditions,

and the procedure utilized in conjunction with the kinetic-catalytic titrations were also used for the variable time determination of nitrilotriacetic acid (NTA). See Chapters II and IV of Part I for the details.

All determinations were carried out at pH ≈ 3.5 in a phosphate-acetate buffer. The manganese concentration was constant depending upon the NTA concentration range being determined. Oxidation of the malachite green cation (MG^+) was followed spectrophotometrically at a wavelength of 620 nm.

For this procedure the following sequence of solutions was used: (1) one ml of malachite green solution (to give an absorbance ≈ 1.0), (2) one ml of the appropriate Mn(II) solution, (3) one ml of NTA solution, (4) five ml of phosphate-acetate buffer, and (5) the final volume in the vessel adjusted to ten ml with purified water.

CHAPTER IV

RESULTS AND DISCUSSION

The periodate oxidation of malachite green cation, MG^+, catalyzed by low concentrations of Mn(II), can be formulated as follows:

$$MG^+ + H_4IO_6^- \xrightarrow{Mn(II)} IO_3^- + \text{colorless products}$$

Upon the addition of low concentrations of nitrilotriacetic acid (NTA) to this system, the catalytic effect of Mn(II) is greatly enhanced. This rate modifying effect of NTA has been used to increase the sensitivity of Mn(II) determination in solution by kinetic methods of analysis (51).

As stated previously, the periodate is added in excess so to create pseudo-zero order dependence with respect to this reactant. A concentrated periodate solution could have been injected instantaneously with a syringe. In this investigation the periodate was still in excess, but it was added at a constant rate of 5.34×10^{-4} ml/second.

Figure 12 shows working curves (in agreement with Equation (11)) for NTA at two concentration ranges. The experimental conditions for NTA at the 10^{-7} M level (curve B) were as follows: reference potential values of $E_L = 0.2429$ volts and $E_U = 0.4027$ volts (0.56 and 0.35 absorbance readings respectively), a constant Mn(II) concentration of 1.0×10^{-5} M, malachite green solution (15 mg/ml), and the periodate was a 10% solution. The only experimental conditions difference for the 10^{-6} M NTA concentration range (curve A) were the Mn(II) concentration, which

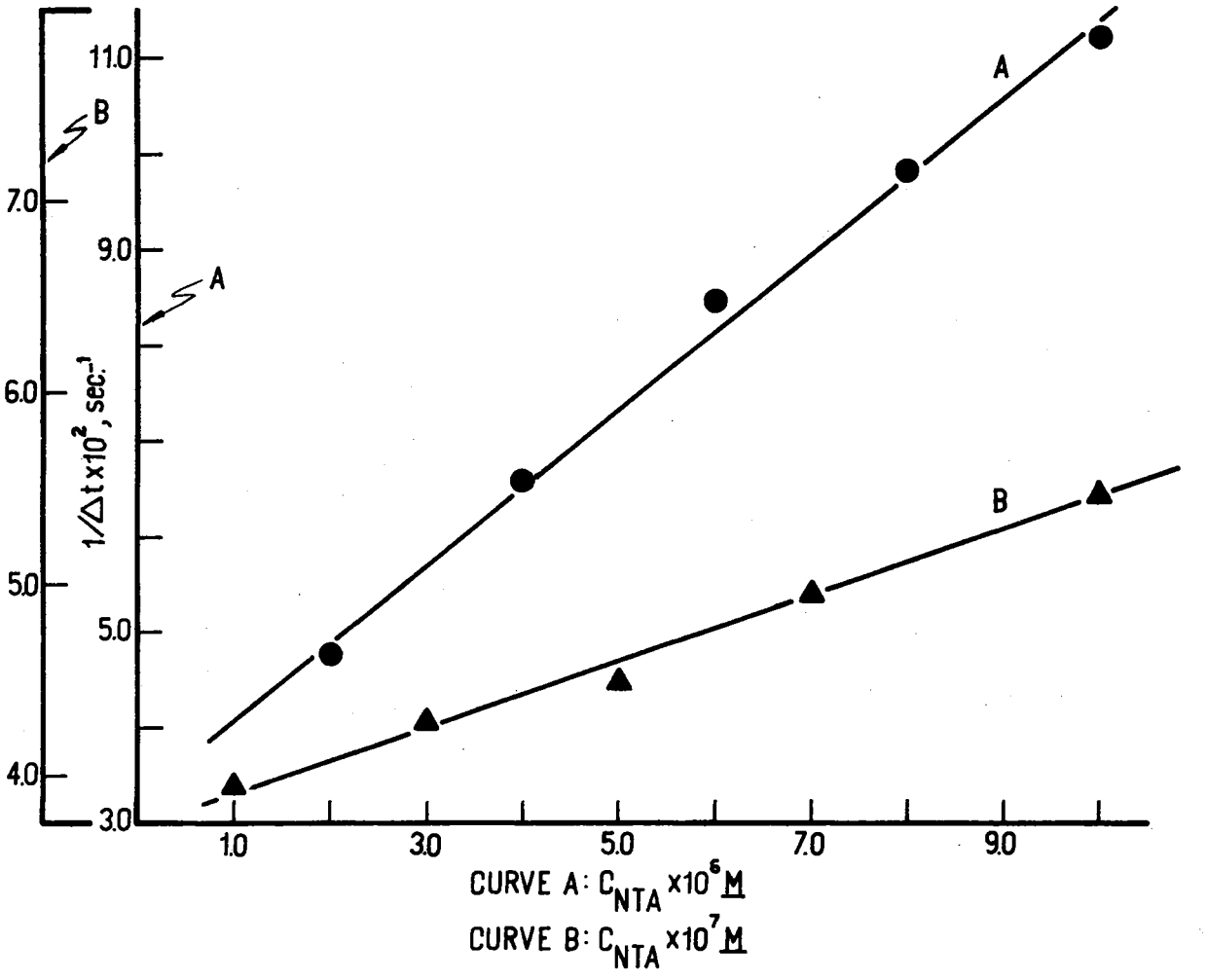

Figure 12. Working Curves for NTA Determination

was changed to 5.0×10^{-6} M, and the reference potentials. Reference potentials were set at $E_L = 0.1447$ volts and $E_U = 0.3346$ volts which correspond to 0.80 and 0.43 absorbance units respectively. A change in Mn(II) concentration was necessary to permit resolution of the recorded elapsed times at the higher NTA concentrations. This in turn allowed an extension of the method to this higher NTA concentration range.

Since $C_{Mn(II)}$ is greater than C_{NTA}, true metal chelate catalysis seems to be indicated in this investigation. If ligand promotion prevailed, the ligand would be destroyed and the rate of the reaction would approach the rate of the metal catalyzed reaction. This was not observed in any of the experimental data obtained.

Even in the case of ligand promotion with $C_{Mn(II)}$ greater than C_{NTA}, this procedure might be of analytical significance. Selection of the reference absorbance values (E_L and E_U) near the beginning of the reaction before the ligand is destroyed could possibly be utilized effectively.

Mn(III) appears to be involved in the catalytic cycle of the manganese catalyzed oxidation of MG^+. At a pH of 3.5 the ratio of complexed Mn(II) (by NTA) to uncomplexed Mn(II) varies approximately from 5.5×10^{-5} to 5.5×10^{-7} (52,53). These values show that most Mn(II) will be as free Mn(II) in solution for the experimental conditions employed to construct the working curves of Figure 12. The same ratio for Mn(III), however, varies from approximately 3.8×10^{6} to 3.8×10^{4} for the same conditions. The formation constant for the Mn(III)-NTA complex used for these calculations was 1.8×10^{20} (54). From these equilibria results, all Mn(III) will be complexed by the NTA at this pH. It therefore seems reasonable to postulate that Mn(III) participates in the catalytic cycle.

If strong complexation of Mn(II) with NTA occurred, the metal catalyst could be subtracted from the catalytic cycle and inhibition by the ligand would be observed.

The total difference in the elapsed times for 2.0×10^{-6} M NTA and 1.0×10^{-5} M NTA was only approximately 12 seconds. Manually measuring the times from a strip chart recorder would again have been virtually impossible. Collection of the elapsed times with the double switch electronic timer was the only reliable and reproducible means available.

This reaction exhibited an induction period that varied from one determination to another. See Table II. Even for duplicate determinations the induction periods were of quite different lengths, but the recorded elapsed times were in good agreement. By selecting the reference absorbance values after the induction period ceases, the variable time procedure can be used where the fixed time method would not be feasible.

The variable time kinetic method of analysis allowed the determination of NTA in the range of 0.2 μg to 2 μg and 2 μg to 20 μg with most relative errors about 1-2% for NTA solutions with no foreign species present. The lower limit of detectability, which is the smallest quantity of material that can be detected with certainty (55), was approximately 0.02 ppm.

Sensitivity may be defined as the ratio of the change in the output signal produced by an increment of the material to be determined to that increment (55). This definition is merely the slope of the working curve with the appropriate units. Sensitivities of these NTA determinations were approximately equal to 0.01 μg^{-1} sec^{-1} at the 10^{-7} M level and 0.004 μg^{-1} sec^{-1} at the 10^{-6} M level.

Seven (out of seven) individual determinations of NTA obtained with

TABLE II

VARIATION OF INDUCTION PERIOD

NTA Concentration ($X10^6$ M)	Length of Induction Period (seconds)	Δt (seconds)
0.00[a]	153	30.5
2.00[a]	153	21.1
2.00[b]	192	33.1
2.00[c]	342	33.2
4.00[a]	189	15.2
4.00[a]	159	15.2
6.00[a]	168	11.8
8.00[a]	184	10.2
10.0[a]	177	8.9

[a] Mn(II) concentration: 5.0×10^{-6} M

[b] Mn(II) concentration: 6.0×10^{-6} M
EDTA concentration: 5.0×10^{-6} M

[c] Mn(II) concentration: 6.0×10^{-6} M
EDTA concentration: 1.0×10^{-6} M

different sets of solutions and on different days gave average values of 2.99 ± 0.13 and 7.05 ± 0.27 for 3.00 and 7.00 micromolar solutions respectively.

Metal ions, some products of degradation of NTA, and other chemical species expected to be present in detergent formulations, natural waters, and industrial waters were investigated as possible interferences for this method. See Table III.

Within certain concentration limits, the determination of NTA by the variable time kinetic method of analysis was relatively free from interferences of foreign species. Sodium tripolyphosphate, calcium ion, dodecylbenzene sodium sulfonate, glycolic acid, and glycine could be tolerated if present in molar concentrations 100 times that of NTA. Sodium lauryl sulfate and aluminum ion did not drastically interfere when present at concentrations ten times that of NTA. From equilibrium calculations, aluminum ion should not have interfered at all (52,53). The observed interference level may be due to impurities in the aluminum salt used in the tests. Iron ion, o-cresol, and iminodiacetic acid did not interfere with the determination if present at the same molar concentration as that of NTA. Since Mn(III)-NTA has a larger formation constant than that of the Fe(III)-NTA complex, the Fe(III) ion should not have interfered either. m-Cresol exhibited some interference, and it would not be expected to do so since o-cresol did not. The o-cresol was 99+% pure and the m-cresol was only 95% pure. The impurities in the m-cresol could possibly have caused this irregular behavior.

Dissolved chlorine proved to be the only serious interference of all foreign species investigated. The MG^{+} was destroyed immediately when added to the solution. Nitrogen was bubbled through the solution

TABLE III

EFFECT OF SOME FOREIGN SPECIES ON THE DETERMINATION OF NTA BY THE VARIABLE TIME PROCEDURE

Species Added	Amount of Species Added (moles/liter)	NTA Added (μg)	NTA Found (μg)	Remarks
$Na_5P_3O_{10}$	5.0×10^{-4}	5.73	6.02	
$Na_5P_3O_{10}$	5.0×10^{-4}	13.4	10.1	
Sodium lauryl sulfate	5.0×10^{-5}	5.73	5.54	
Sodium lauryl sulfate	5.0×10^{-5}	13.4	12.6	
Sodium lauryl sulfate	5.0×10^{-4}	13.4	0.00	No catalysis
$Ca(ClO_4)\cdot 6H_2O$	5.0×10^{-4}	5.73	4.97	
$Ca(ClO_4)\cdot 6H_2O$	5.0×10^{-4}	13.4	13.0	
Dodecylbenzene sodium sulfonate	5.0×10^{-4}	5.73	5.73	
Dodecylbenzene sodium sulfonate	5.0×10^{-4}	13.4	14.2	
$Fe(ClO_4)_3$	3.5×10^{-6}	5.73	4.20	
$Fe(ClO_4)_3$	3.5×10^{-6}	13.4	10.9	
$Fe(ClO_4)_3$	3.5×10^{-5}	5.73	0.00	Same Δt as blank
$Fe(ClO_4)_3$	3.5×10^{-5}	13.4	0.00	Same Δt as blank
o-Cresol	5.0×10^{-6}	5.73	6.11	
o-Cresol	5.0×10^{-6}	13.4	14.0	
o-Cresol	5.0×10^{-5}	5.73	0.00	$\Delta t >$ blank
m-Cresol	4.0×10^{-6}	5.73	4.01	Absorbance had to be adjusted to ≈1 with Mg^{+}
m-Cresol	4.0×10^{-6}	13.4	7.66	

TABLE III (Continued)

Species Added	Amount of Species Added (moles/liter)	NTA Added (μg)	NTA Found (μg)	Remarks
Iminodiacetic acid	5.0×10^{-6}	5.73	4.68	
Iminodiacetic acid	5.0×10^{-6}	13.4	13.4	
Glycolic acid	5.0×10^{-6}	5.73	5.35	
Glycolic acid	5.0×10^{-6}	13.4	12.6	
Glycolic acid	5.0×10^{-4}	13.4	13.8	
Glycine	5.0×10^{-6}	5.73	5.73	
Glycine	5.0×10^{-6}	13.4	13.4	
Glycine	5.0×10^{-4}	5.73	5.73	
$AlNa(SO_4)_2 \cdot 12H_2O$	5.0×10^{-6}	5.73	8.40	
$AlNa(SO_4)_2 \cdot 12H_2O$	5.0×10^{-6}	13.4	17.0	
$AlNa(SO_4)_2 \cdot 12H_2O$	5.0×10^{-5}	5.73	3.06	
$AlNa(SO_4)_2 \cdot 12H_2O$	5.0×10^{-5}	13.4	10.3	
Dissolved chlorine	Saturated solution	5.73	0.00	MG^+ destroyed when added
Dissolved chlorine	Saturated solution	13.4	0.00	MG^+ destroyed when added

to expel the chlorine, but the MG^+ was still destroyed. This was thought to be caused by the hypochlorite ion which is formed when a solution is saturated with chlorine.

Hydrazine hydrochloride was used to try to reduce the hypochlorite ion to chloride ion. The MG^+ was not destroyed when added, but upon addition of the periodate to the solution nitrogen gas was liberated.

Ascorbic acid was utilized as a reducing agent and the MG^+ was not destroyed when added. Upon the addition of one drop of periodate, the MG^+ changed from its normal green color to a dark brownish green.

Hydrogen peroxide was tried in a like manner and a gas was given off near the end of the determination. These bubbles collected on the walls of the flow cell and caused irregular results.

Sodium nitrite was employed as a reducing agent for the hypochlorite ion. The MG^+ was not destroyed, but the reaction was inhibited tremendously as two syringes of periodate were added and the oxidation of MG^+ never did occur. The same thing occurred when ammonia or sodium sulfite were used. Magnesium metal was tried, but it proved ineffective due to complete inhibition of the reaction.

Figures 13 and 14 present working curves for the determination of NTA in the presence of EDTA following the same procedure. Mn(II) and EDTA concentrations were 1.5×10^{-5} M and 1.0×10^{-5} M respectively for both determinations. A 10% periodate solution was again added at a constant rate and the malachite green solution was the same as was used for the determination of NTA without EDTA present. Figure 13 had a shorter elapsed time collection interval which improved the sensitivity of determining NTA in the presence of a known amount of EDTA. By selecting the appropriate amount of Mn(II), from these working curves it can be

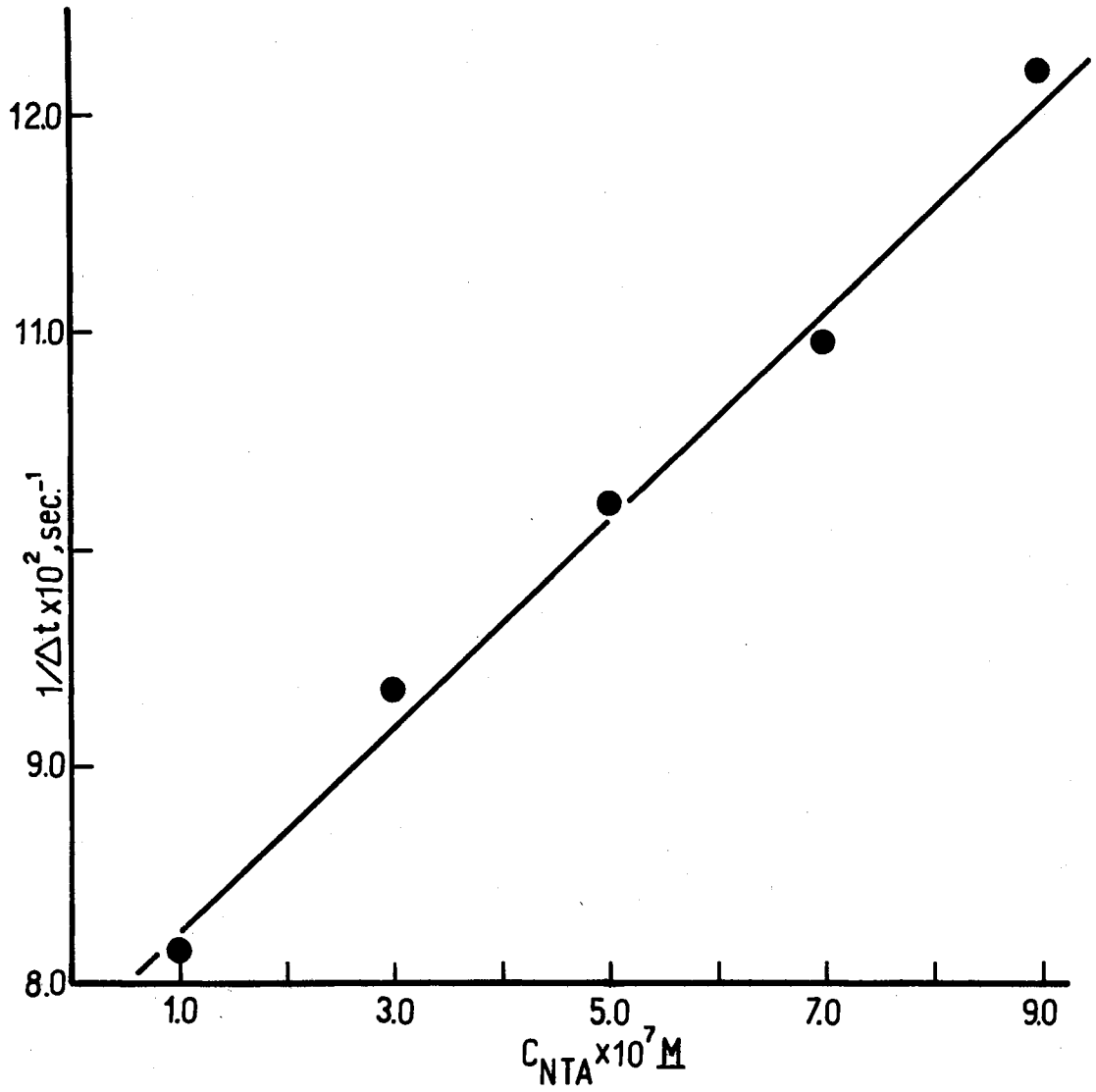

Figure 13. Working Curve for NTA Determination in the Presence of 1.0×10^{-5} M EDTA

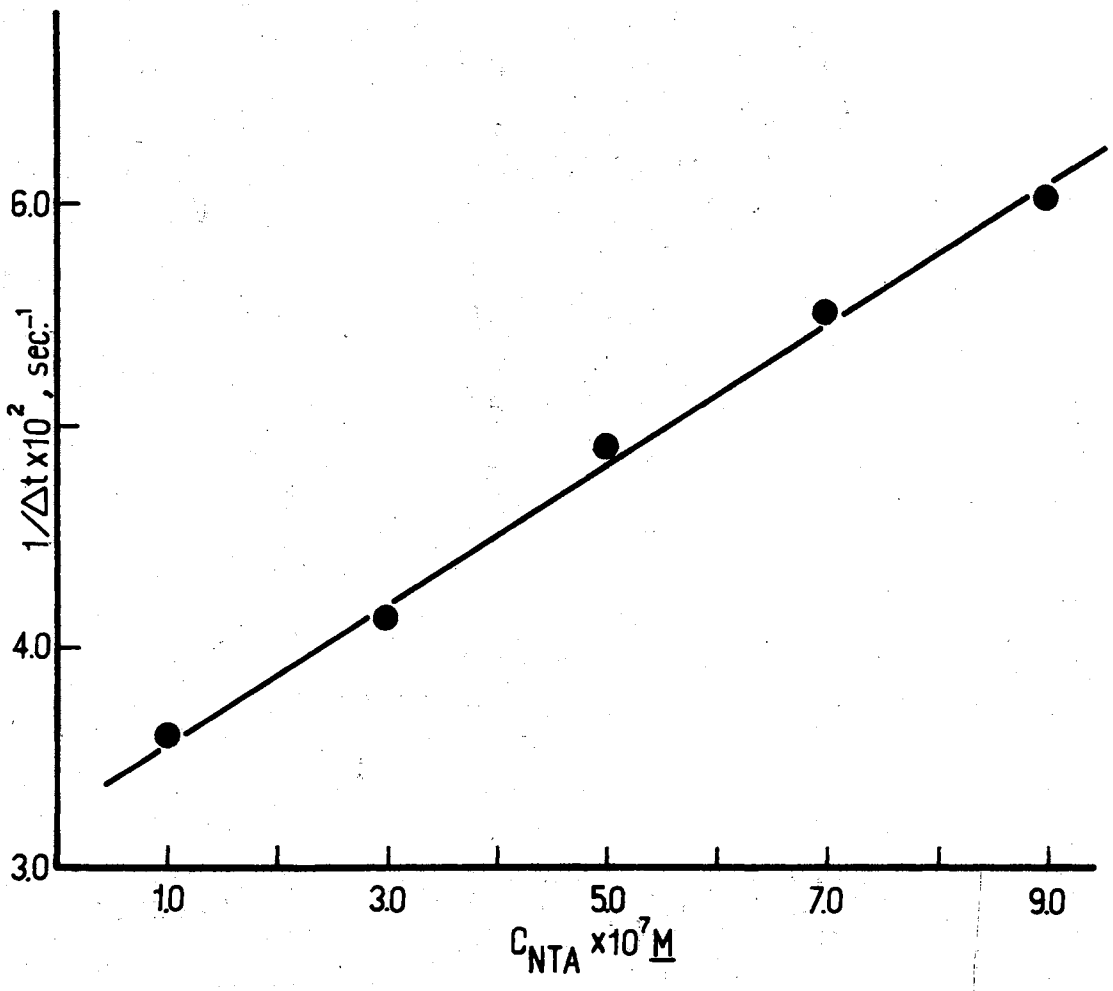

Figure 14. Working Curve for NTA Determination in the Presence of 1.0×10^{-5} M EDTA

inferred that this procedure could be effectively utilized for the determination of NTA in samples of aminopolycarboxylic acid (or other complexing agents) that inhibit the catalytic effect of manganese.

The amount of Mn(II) that was required in these previous determinations of NTA was, of course, dependent upon the amount of EDTA present in the sample. This procedure, however, can be applied if the amount of EDTA is approximately known. To overcome this prerequisite, the standard addition method was applied (56). The method of standard addition depends upon the calibration curve passing through the origin of the possibility of correcting for the blank. From Equation (11), it can be seen that the intercept would not necessarily pass through the origin because k_c and $[C]_o$ could be large when compared to $(k'_c - k_c)$. The $1/\Delta t$ value for the blank solution (no NTA) cannot be experimentally determined because the free metal catalyst concentration, [C], depends upon the concentration of EDTA as well as the EDTA/NTA ratio. Since they are unknown in real samples, the correction for the blank will vary in an unpredictable manner and the standard addition method thus fails in conjunction with this procedure. If relatively large errors can be tolerated, however, a rough estimation of the possible NTA content in a sample could be estimated.

Another method that was tried in order to determine NTA in the presence of EDTA was that of adding Mn(II) as a titrant instead of periodate. The EDTA should inhibit the reaction until all of the EDTA is complexed by the Mn(II). Then the Mn(II) could participate in the catalytic path and NTA would be determined in a manner analagous to the original procedure. Results obtained following this procedure were erratic and were possibly due to the buildup of manganese. Once the reaction was initi-

ated, the NTA effect was masked by the high manganese concentration in the reaction vessel.

This variable time kinetic method of analysis allows selective detection and determination of ppm and fractions of ppm of NTA. The relatively low pH at which the method is used reduces the possibility of interferences from other metal ions and complexing agents. Monitoring and repetitive analysis should be easily adaptable to this procedure.

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