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A SIMPLE COMPUTER CIRCUIT FOR AUTOMATIC SPECTROPHOTOMETRIC ANALYSIS OF BINARY MIXTURES BY DIFFERENTIAL REACTION RATES

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Summary—A simple analogue computer circuit, for application with a continuous reading spectrophotometer to give automatic analysis of binary mixtures of closely related substances using a differential reaction rate technique, is described. The circuit solves the simultaneous equations of the Method of Proportional Equations for the concentrations of the components in the mixture. The method is useful for first- or pseudo-first order competitive reactions. A timing circuit automatically supplies the absorbance (converted as described from the transmittance) of the reacting solution at two chosen times during the reaction, to the computer. The output voltages are adjusted within the circuit to read directly in units of concentration.

INTRODUCTION

In recent years several analytical techniques based on differential reaction rates have been devised for the *in situ* simultaneous quantitative determination of mixtures of closely related substances.[1–11] All the techniques devised require a rather laborious graphical[5] or mathematical[1] treatment of the data in order to arrive at the concentrations of the unknowns of interest. This paper describes the circuit of an automatic read-out system for the *Method of Proportional Equations*. It can be attached to virtually any continuous reading spectrophotometer that gives an electrical output signal proportional to the transmittance of the sample solution. The simultaneous equations are solved by a simple analogue computer circuit.

PRINCIPLES OF AUTOMATIC READ-OUT CIRCUIT

The *Method of Proportional Equations* can be used for the simultaneous analysis of mixtures of closely related substances if a reagent R can be made to react under pseudo first order conditions with each of the n components $A, B, \ldots, N$ of the mixture at different rates, $K_A, K_B, \ldots, K_N$ to form a common product, O, or different products yielding a similar instrument response.

The circuit described in this paper was designed for the analysis of a two component mixture. However, by following the principles given below it can easily be extended to mixtures of more than two components. The reaction product(s) that are formed are assumed to absorb light at the same wavelength (if two products result, the fact that they might have different extinction coefficients at the wavelength used does not effect the validity of the method[1,6]) and to follow Beer's law. The proportional

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equations for two unknowns have the form:

$$P_1 = K_{A_1}[A]_0 + K_{B_1}[B]_0 \quad (1)$$

$$P_2 = K_{A_2}[A]_0 + K_{B_2}[B]_0 \quad (2)$$

where P_1 and P_2 are the experimentally measured parameters which are proportional to the absorbance of the reaction mixture at times t_1 and t_2 during the reaction, K_{A_1}, K_{B_1}, K_{A_2}, and K_{B_2} are the proportionality constants, and $[A]_0$ and $[B]_0$ are the initial concentrations of the species to be analysed [see references 1 and 6 for the derivation of equations (1) and (2)].

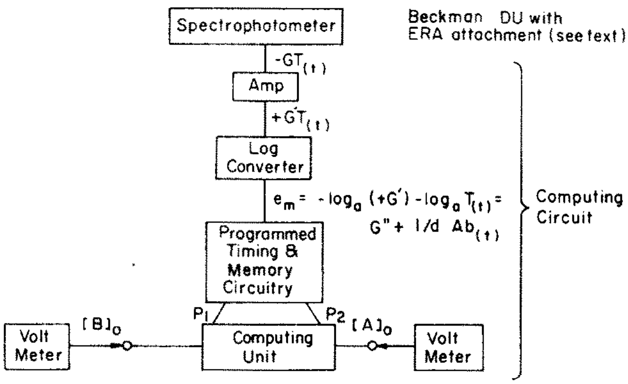

FIG. 1.—Block diagram of automatic read-out apparatus.

A block diagram of the complete analytical system is shown in Fig. 1. The spectrophotometer used in this work was a Beckman DU equipped with a Beckman Energy Recording Adapter (ERA). The ERA converts the phototube current to a voltage suitable for the input of a recorder. [Any recording spectrophotometer can be used with the read-out circuit described, provided that the instrument can be operated at a fixed wavelength. The instrument's output (the input to the spectrophotometer's recorder) is fed directly into the read-out circuit.] The output of the ERA unit, $-GT(t)$, is proportional to the transmission, $T(t)$, of the sample solution at the selected wavelength at any time during the reaction. The proportionality constant is G. Voltages proportional to the absorbance are needed for computation: the logarithm of the output of the ERA must be taken. First, however, the voltage, $-GT(t)$, is fed into a high input impedence amplifier which serves a dual purpose: its high input impedence prevents loading of the ERA output, and it amplifies the original signal $-GT(t)$ to a new signal $+G'T(t)$ which is in the range (1–10 V) for proper operation of the log circuit. By its nature, the amplifier inverts the sign of the input.[12] The device used to perform the logarithmic operation on the input signal was a slightly modified version of the circuit described by Savant and Howard.[13] The response of the circuit was directly logarithmic for the above input voltage range with a 1-V input corresponding to a 0-V output. The output of the log circuit is thus:

$$e_m = -\log_a [G'T(t)] = -\log_a G' - \log_a T(t). = +G'' - \log_a T(t) \quad (3)$$

where G'' is the constant $-\log_a G'$.

The absorbance, $Ab(t)$, of a system is defined in terms of transmission

$$Ab(t) \equiv -\log_{10} T(t) = d \log_a T(t) \tag{4}$$

Substituting equation (4) into (3) gives the output of the log circuit in terms of the absorbance and the constants G'' and $1/d$

$$e_m = +G'' + 1/d\, Ab(t) \tag{5}$$

At two preselected times, t_1 and t_2, during the reaction, a programmed timer feeds the signal e_m into memory units. In the memory units, the sign of e_m is inverted; thus, two voltages $-P_1 - \alpha G''$ and $-P_2 - \alpha G''$ (α is a constant introduced by the memory unit) are available for computation of $[A]_0$ and $[B]_0$ by a simple computer circuit at any time after t_2. For detailed descriptions of the memory and computer circuits see below.

Two operational amplifiers employed as conventional integrating circuits[12,14] were used as the memory units (see Fig. 2). The voltage e_m entering the integrator circuit is of the form in equation (5). At time t_1, the switching circuit applies $e_m(t_1)$ to the integrator for a time Δt_1 that is short in comparison to the reaction time, t_1. The output of the integrator is then:

$$e_{m_1}\ \text{out} = -\frac{1}{RC}\int_{t_1}^{t_1+\Delta t_1} e_m(t)\, dt = -\frac{1}{RC}\int_{t_1}^{t_1+\Delta t_1} \left[G'' + \frac{1}{d} Ab(t)\right] dt \tag{6}$$

Because Δt_1 is short, $e_m(t_1)$ can be considered constant during the interval and to have a value $e_m(t_1)$. (R is the value of the resistance and C the capacitance of the elements of the integrator circuit and e_{m_1} is the output of the memory circuit.) Thus,

$$e_{m_1}\ \text{out} = -\frac{1}{RC}\int_{t_1}^{t_1+\Delta t_1} e_m(t)\, dt = -\frac{1}{RC}[G'' + \frac{1}{d} Ab(t_1)]\Delta t_1 \tag{7}$$

$$= -\frac{\Delta t_1}{RC} G'' - \frac{\Delta t_1}{RC}\frac{Ab}{d}(t_1)$$

$$= -\alpha G'' - \frac{\alpha}{d} Ab(t_1) \tag{8}$$

where α = a constant $= \Delta t_1/RC$. Note that the sign of the voltage of equation (5) has been inverted.[12] At time t_2 the programmed timer feeds the signal $e_m(t_2)$ into the second integrator. By the same arguments as above the signal output of the second memory unit is:

$$e_{m_2}\ \text{out} = -\alpha' G'' - \frac{\alpha'}{d} Ab(t_2) \tag{9}$$

The two integrators hold (as a memory) e_{m_1} out and e_{m_2} out, respectively, after the input signals are applied.[12] If $\Delta t_1 = \Delta t_2$ and the RC constants of the integrators are identical, $\alpha = \alpha'$. The two signals in the memory are:

$$\left.\begin{aligned} e_{m_1}\ \text{out} &= -\alpha G'' - \frac{\alpha}{d} Ab(t_1) = -\alpha G'' - P_1 \\ e_{m_2}\ \text{out} &= -\alpha G'' - \frac{\alpha}{d} Ab(t_2) = -\alpha G'' - P_2 \end{aligned}\right\} \tag{10}$$

The use of integrator circuits for the memories allows a small but finite current to be drawn from them for computing without effecting the values e_{m_1} out and e_{m_2} out.[12]

The voltages in the memory are proportional to the absorbance at t_1 and t_2 [equations (10)] but contain the additive constant potential $-\alpha G''$. At a finite time, 1 min after t_2, the programmed timer then applies e_{m_1} out and e_{m_2} out to the two operational amplifiers of the computing circuit. (See below for a detailed discussion of the timer.) The computing circuit (see Fig. 2) used is a standard analogue circuit for solving a system of simultaneous equations.[11] Simultaneously, the timing circuit applies a voltage equal to $+\alpha G''$ to each amplifier. The circuit is designed so that the $+\alpha G''$ voltage is added to both signals, e_{m_1} out and e_{m_2} out. The result is an effective input to the computer of $-P_1 = \left[-\frac{\alpha}{d}Ab(t_1)\right]$ and $-P_2 = \left[-\frac{\alpha}{d}Ab(t_2)\right]$. These voltages are thus directly proportional to the concentrations of products of the reactions at the times t_1 and t_2, and are exactly the voltages necessary to solve equations (1) and (2) for $[A]_0$ and $[B]_0$. The input and feedback impedences of the computer network are chosen to make the output of the computer read directly in terms of the concentrations of $[A]_0$ and $[B]_0$. No further calculation is necessary. (For a detailed discussion of the computing circuit, see below.) It is a simple matter to connect two voltmeters to read $[A]_0$ and $[B]_0$. Digital voltmeters, such as the Electro Instruments (San Diego, California, U.S.A.) Model 4000 Digital Voltmeter, are recommended to give numerical display of the read-out.

It should be noted that although the above circuit is constructed for a spectrophotometric method of following the reaction, any method that gives an output signal directly proportional to the concentration of product(s) can also be used. In such a case, the logarithmic circuit is omitted and the amplified signal applied directly to the timer-memory circuit.

EXPERIMENTAL

Construction and operation details of each circuit in the automatic read-out system are given below.

Amplifier circuit

This circuit consists of two parts, (i) a standard type high input impedance ($\sim 10^{13}\,\Omega$) voltage follower F which prevents loading of the spectrophotometer output,[12] and (ii) a variable gain amplifier (gain of $-$G) which is capable of amplifying the signal by a factor of -1, -10, -10^2 and -10^3. (The sign inversion is inherent in analogue circuits of this type.[14]) These circuits are standard and the details of their operation and associated equipment (power supply, bias, *etc.*) are found in references 12 and 14.

Logarithmic circuit

The logarithmic circuit is essentially the same as that described by Savant and Howard.[13] The 350 Ω and 3·5 K potentiometer are used to bias the tube so that it operates on the logarithmic portion of its characteristic curve for the 1–10 V input range. The 10 K and 250 K potentiometer are then adjusted to make the output read 0 V with a 1-V input. It was found that the rise-time response of this circuit on a 1-V instantaneous change input was about 10 sec. This is well within the time required to follow most reactions for which this technique would be employed,[1,15] but must be considered when the unit is used with fast reactions. Considerable current is drawn from the -6 V bias supply because there is only 350 Ω to ground at this point. If a standard type "C" battery is used, there is a noticeable change in the circuits' characteristics after continuous operation for 6 hr. This can be reduced by using an automobile battery or a commercial transistorised low voltage power supply instead. The ± 300 V d.c. operating voltage of the circuit is supplied by the same power supply that runs the operational amplifiers (a Philbrick R-100 B $\pm$ 300 V d.c. power supply was used[14]). The adjustable base resistor of this circuit[13] is used to adjust the logarithmic base to a convenient value to give a slope of about 1 on a semi-log plot of output *vs.* input. A value of 10 Meg was used in this work. Other satisfactory logarithmic circuits based on the logarithmic characteristics of certain transistors are described in the literature.[16]

Programmed sequence timing and memory circuit

As stated before, the memory circuits are simply two conventional analogue computer operational amplifiers (Philbrick K2-W, K2-P stabilised units were used[14]) connected as integrators[13,14] (Fig. 2). The two variable resistors R_6 in the integrator circuits are used to equalise the RC time constants of the integrators [see equations (7)–(9)]. The two identical bridge circuits (R_8, R_9, R_{10} and R_{11}) are connected to the summing points[14] of the two integrators. They supply a small current (adjustable by R_{10}) to compensate for leakage in the system (amplifier grid current, capacitor leakage, *etc.*).[12]

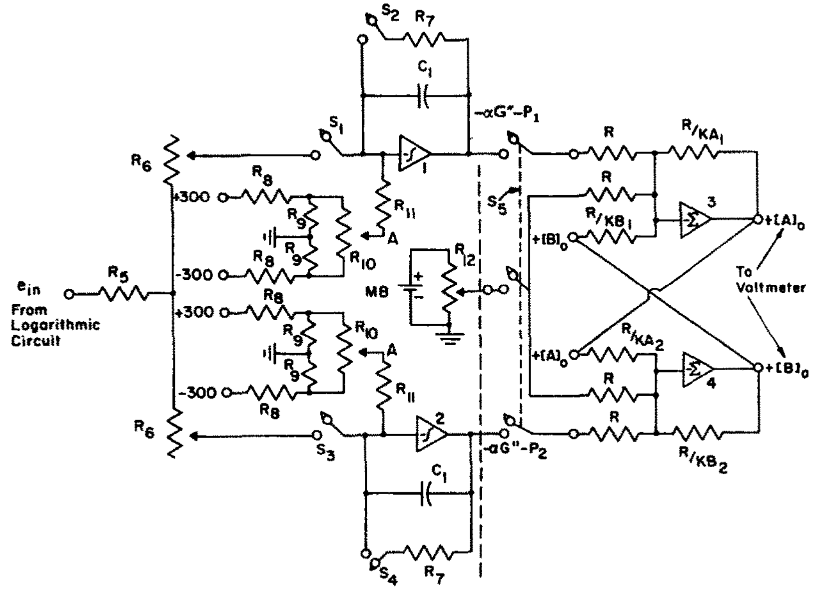

FIG. 2.—Memory and computing circuits:
R = 100 k to 10 Meg (see text)
R_5 = 7 M 1%
R_6 = 1M—Used to equalise RC time constants of integrators
R_7 = 500 Ω
C_1 = 1·0 Mfd R_{10} = 500Ω MB = Mercury battery
R_8 = 1 M 1% R_{11} = 10 M of necessary
R_9 = 39 Ω R_{12} = 1K, 2W magnitude.

The timing circuit, shown in Figs. 2 and 3, operates as follows: Switches S_8, S_9 and S_{10} are microswitches that are activated by a synchronous timer. S_8 can also be activated by a latching relay (Fig. 3), R built into the timer and controlled by S_6 (this timer is an Industrial Timer Corporation Model RC-8 unit). Closing switch S_6 starts the timer motor by activating S_8. [Note. Fig. 3 shows the circuit just after the cycle has been started and before any signal has been applied to the memory]. S_8 then stays in the position shown in Fig. 3 until the end of the cycle when it automatically opens and stops the motor. At a time t_1 during the reaction, S_9 is closed by the timer cam for a time Δt_1 that is *small* (less than 1%) compared to the over-all reaction time. This accomplishes two operations: (i) It opens the latching relay S_2 so that the short circuit (discharge path) though R_7 around capacitor C_1 (see Fig. 2) is opened, which permits C_1 to then store a potential [S_2 remains latched in this position for the rest of the cycle until manually reset by closing S_7 momentarily (described later)]. (ii) Simultaneously, S_1, which applies the output of the logarithmic circuit at t_1 into the integrator is closed. It opens again after a time Δt_1 when S_9 opens again (timer activated). Thus $G'' + \frac{1}{d} Ab\,(t_1)$ is applied to, and its integral is stored on, integrator 1. At time t_2, the timer closes S_{10} for a time $\Delta t_2 = \Delta t_1$. Relays S_3 and S_4 operate similarly to S_1 and S_2 in controlling the sequence of events in the second integrator circuit. Then, at any time $t > t_2$, the output of integrator 1 is $-\alpha G'' - P_1$ and the output of integrator 2 is $-\alpha G'' - P_2$. At a time $t > t_2$, dependent on the duration of the timer cycle, the timer automatically turns itself off. Switch S_8 goes to the upper position [position (1)] in Fig. 3. This activates relay S_5. This relay then applies $-\alpha G'' - P_1$, $-\alpha G'' - P_2$ and $+\alpha G''$ into the inputs of the computing circuit [see Fig. 2 and equation (10)]. The computer then develops $[A_0]$ and $[B_0]$ as output voltages and will hold these "answers." (The computing process is described below). To reset the latching relays, S_2, S_4 and S_5, and thus erase the memory in preparation for another run, S_7 is closed momentarily. However, it will be noticed that if S_8 is in the upper position [position (1) in Fig. 3], S_5 will get a signal to activate again. Thus, the proper reset sequence is the following: close S_6 momentarily to activate relay R and thus S_8. The timer motor is now running [S_8 in position (2)]. Immediately close S_7 momentarily to reset S_2, S_4 and S_5. (If it is desired to erase the memories without reactivating the cycle, just close S_7, but recall that S_5 will *not* be reset.)

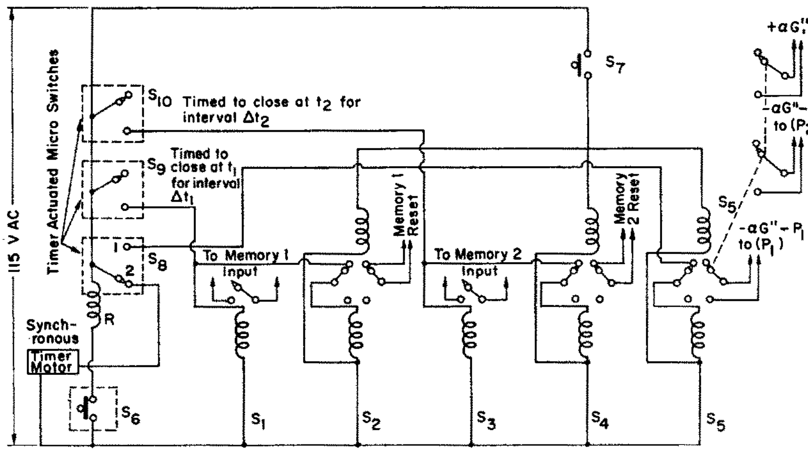

FIG. 3.—Programmed Timing Circuit (controls the sequence of the activation of the switches in the memory and computing circuits; switch numbers are the same as in Fig. 2):
Timer—Industrial Timer Corp., Model RC-8;
S_1 and S_3—Potter-Brumfield Relay—Model #KA11AY 115 V a.c.;
S_2, S_4 + S_5—Potter-Brumfield Latching Relay—Model #KB17AY 115 V a.c., 4PDT.

Switches S_8, S_9 and S_{10} are cam operated and t_1, Δt_1, t_2 and Δt_2 are easily preset to any desired value. The desired duration of the full cycle will depend on the speed of the reaction that is being followed. The length of the timer cycle can be adjusted by simply changing the synchronous motor gear ratios. Different sets of gears are commercially available that can vary the cycle from 10 sec to several hours.

Computing circuit

The computing circuit consists of two summing operational amplifiers[12,14] (Fig. 2). The principles governing the application of summing amplifiers in solving simultaneous equations can be easily shown by rearranging the proportional equations (1) and (2) in the form

$$[A]_0 = \frac{P_1}{K_{A_1}} - \frac{K_{B_1}}{K_{A_1}}[B]_0 \tag{11}$$

$$[B]_0 = \frac{P_2}{K_{B_2}} - \frac{K_{A_2}}{K_{B_2}}[A]_0 \tag{12}$$

The memory circuits have stored the voltages $-\alpha G'' - P_1$ and $-\alpha G'' - P_2$, which can be substituted as input voltages (e_{in_1} and e_{in_2}) along with the voltage $+\alpha G''$ (e_{in_3}) in the expression describing the operation of a summing amplifier[14] (the value of the feedback resistor, R_f, is any convenient value such that all the resistance values fall between 10 K and 20 M). One obtains:

$$\begin{aligned} e_{out} &= -(-\alpha G'' - P_1)\frac{1}{K_{A_1}} - \alpha G''\frac{1}{K_{A_1}} - (+[B]_0)\frac{K_{B_1}}{K_{A_1}} \\ &= +\frac{P_1}{K_{A_1}} - [B]_0\frac{K_{B_1}}{K_{A_1}} \end{aligned} \tag{13}$$

This is the expression for $+[A]_0$ of equation (11). A similar circuit can be used to calculate $+[B]_0$. [The voltage $+\alpha G''$ is supplied by means of a mercury battery MB which can be varied from 0 to about 20 V and it is adjusted precisely by the potentiometer (10 turn Helipot) R_{12}.] Of course, the values, $+[B]_0$ and $+[A]_0$, which are the solutions sought, are not available as such for substitution into the right hand side of equations (11) and (12). However, if the output of amplifier 3 is fed back into the $+[A]_0$ input of amplifier 4 and the output of 4 fed into the $+[B]_0$ input of amplifier 3, when the potentials $-\alpha G'' - P_1$, $-\alpha G'' - P_2$ and $+\alpha G''$ are applied to the respective inputs (see Fig. 2), the circuit will rapidly come to steady state condition with $+[A]_0$ and $+[B]_0$ reading at the outputs.[1,17] This type of *solution feedback* is the basic principle of all analogue computation.[1,12,14,17]

The values of K_{A_1}, K_{B_1}, K_{A_2} and K_{B_2} are experimentally determined by reacting a solution of pure A and then pure B and measuring P_1 and P_2 for each at times t_1 and t_2. The values of K_{A_1}, *etc.*, are then calculated simply from:

$$K_{A_1} = P_{A_1}/[A]_0$$

$$K_{B_1} = P_{B_1}/[B]_0 \qquad (14)$$

$$\vdots$$

etc.

In order to determine the accuracy and precision of the automatic read-out unit, a large number of simulated reaction rate curves were applied to this unit by means of an electronic function generator. These simulated rate curves were constructed from theory using several different cases. (Different rate constants and ratios of $[A]_0/[B]_0$) were used, rather than actual experimental rate curve responses from the spectrophotometer in the evaluation of the automatic unit in order to eliminate all source of variation of parameters not directly introduced by the read-out unit. Thus, no error resulting from spectrophotometer drift, temperature change of the reacting solution, *etc.*, complicated the comparison of the hand calculated and automatic read-out results, and the accuracy and precision of the automatic read-out unit is obtained exactly.) It was found that the results obtained by the automatic read-out unit had an accuracy of better than $\pm 2\%$ when compared with the hand calculated (theoretical) results and a precision (standard deviation) of less than $\pm 2\%$, when care is taken in measuring the proportionality constants, K's, and in adjusting the instrument.

The increased speed over hand calculation of determining the values of the experimental proportionality constants of the system as well as the analysis result make this unit very useful when large numbers of analyses are being made with this kinetic method. The computing unit would be even more useful when the mixtures contained three or four components because the calculation becomes very tedious in these cases.

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Zusammenfassung—Eine einfache Analogrechnerschaltung wird beschrieben, die mit Hilfe eines kontinuierlich anzeigenden Spektralphotometers eine automatische Analyse binärer Mischungen nahe verwandter Substanzen liefert, wobei eine Technik verwendet wird, die sich auf Reaktionsgeschwindigkeitsunterschiede gründet. Die Schaltung löst die simultanen Gleichungen der Methode der proportionalen Gleichungen für die Konzentrationen der Bestandteile in der Mischung. Die Methode ist von Nutzen bei Konkurrenzreaktionen erster oder pseudoerster Ordnung. Eine Zeitgeberschaltung gibt die Extinktion (die auf bekannte Weise aus der Durchlässigkeit erhalten wird) der reagierenden Lösung zu zwei wählbaren Zeiten während der Reaktion automatisch in den Rechner. Die Ausgangsspannungen werden in der Schaltung so justiert, daß sie direkt Konzentrationseinheiten angeben.

Résumé—On décrit un circuit calculateur analogique simple, en liaison avec un spectrophotomètre à lecture continue, pour l'analyse automatique de mélanges binaires de substances étroitement apparentées, par l'emploi d'une technique de vitesses de réaction différentielles. Le circuit résout les équations simultanées de la méthode des équations proportionnelles, pour les concentrations des composants du mélange. La méthode est utile pour les réactions concurrentes d'ordre un ou pseudo-un. Un circuit chronométreur fournit automatiquement au calculateur l'absorption (à partir, ainsi qu'il est décrit, de la transmission) de la solution réagissante à deux instants choisis durant la réaction. Les voltages à la sortie sont ajustés dans le circuit de façon à permettre la lecture directement en unités de concentration.

REFERENCES

[1] H. B. Mark, Jr., L. J. Papa and C. N. Reilley, *Advances in Analytical Chemistry and Instrumentation*, Vol. II. Interscience Publishers, New York, 1963, p. 255.
[2] T. S. Lee and I. M. Kolthoff, *Ann. N.Y. Acad. Sci.*, 1954, **40**, 70.
[3] J. D. Roberts and C. Regan, *Analyt. Chem.*, 1952, **24**, 360.
[4] L. J. Papa, J. H. Patterson, H. B. Mark, Jr. and C. W. Reilley, *ibid.*, 1963, **35**, 1889.
[5] S. Siggia and J. G. Hanna, *ibid.*, 1961, **33**, 896.
[6] R. G. Garmon and C. N. Reilley, *ibid.*, 1962, **34**, 600.
[7] L. J. Papa, H. B. Mark, Jr. and C. N. Reilley, *ibid.*, 1962, **34**, 1513.
[8] H. B. Mark, Jr., *ibid.*, 1964, **36**, 1668.
[9] L. J. Papa, H. B. Mark, Jr. and C. N. Reilley, *ibid.*, 1962, **34**, 1443.
[10] J. G. Hanna and S. Siggia, *ibid.*, 1962, **34**, 547.
[11] J. G. Hanna and S. Siggia, *J. Polymer Sci.*, 1962, **56**, 297.
[12] D. D. DeFord, Division of Anal. Chem., 133rd Meeting, ACS, San Francisco, 1958; G. A. Philbrick Researches, Inc., Applications Bulletin 12-19-57, 127 Clarendon Street, Boston 16, Mass.
[13] C. J. Savant, Jr. and R. C. Howard, *Electronics*, 1954, **27**, 144.
[14] G. A. Philbrick Researches, Inc., *Applications Manual for Computing Amplifiers*, 127 Clarendon Street, Boston 16, Mass. (for an extended review of operational Amplifiers: see Operational Amplifier Symposium, *Analyt. Chem.*, 1963, **35**, 1770–1833).
[15] L. J. Papa, H. B. Mark, Jr. and C. N. Reilley, unpublished results.
[16] W. L. Paterson, *Rev. Sci. Instr.*, 1963, **34**, 1311.
[17] C. N. Reilley, *J. Chem. Educ.*, 1962, **39**, A933.