Simplified Automatic Determination of *In Vitro* Drug Dissolution Rates Using Direct Concentration Recording

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Abstract □ A system is described which permits automatic recording of dissolution rates directly in terms of percentage dissolution *versus* time without the need for an intermediate absorbance–time plot.

Keyphrases □ Automated system—*in vitro* dissolution rates □ Dissolution rates—automated procedure □ Concentration converter—absorbance–time curves □ UV spectrophotometry—analysis

During the past decade, studies of dissolution behavior of tablets and capsules have gained in importance because of the growing awareness of differences among products containing the same drug. The terms "therapeutic equivalency" and "physiological availability" have been employed to highlight the fact that different formulations of the same drug may affect the rate and extent of absorption after oral administration. Schroeter *et al.* (1), Levy *et al.* (2), and Nelson (3) related *in vitro* dissolution behavior of drugs to their *in vivo* absorption.

Various methods for determining *in vitro* dissolution rates were recently reviewed (4). Of the several analytical methods for monitoring drug concentration during a dissolution study, spectrophotometry remains the simplest and most rapid. Determination of dissolution characteristics of a drug preparation requires that measurements be made on a large number of samples. Steadily increasing demands for data on dissolution rates make some degree of automation of obvious benefit.

APPARATUS

The dissolution apparatus employed in this laboratory was previously described (5). The dissolution vessel is a V-shaped, transparent, plastic chamber which oscillates about its center. The frequency of oscillation may be changed by means of a variable speed motor. The rate of oscillation chosen should furnish sufficient agitation to maintain homogeneity in the chamber without imparting unusual mechanical stress to the capsule or tablet. The dissolution medium is maintained at 37° by partially immersing the chamber in a constant-temperature bath.

Fluid is pumped from the dissolution chamber using a Varistaltic[1] pump through polyethylene and Tygon[2] tubing and through a 0.2-cm. silica flowcell in the sample beam of a Beckman model DB spectrophotometer. A glass-wool filter in the pump intake line prevents undissolved solids from entering the cell.

The output signal from the spectrophotometer is 0–100 mv. for 0–100% T (transmittance). This signal is conditioned and recorded using a Beckman model 131902 concentration converter and 100-mv. potentiometric strip chart recorder.

[1] Manostat Corp., New York, N. Y.
[2] U. S. Stoneware Co., Akron, Ohio.

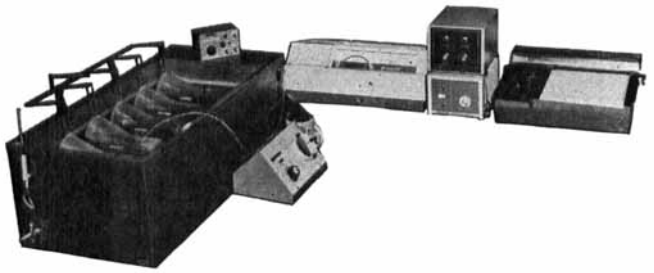

Figure 1—In vitro *drug dissolution apparatus.*

The output signal from the concentration converter can be presented as absorbance over a variety of ranges, and concentration can be presented over a single, adjustable range. Figure 1 is a photograph of the assembled system, and Fig. 2 is a flow diagram of the apparatus.

Previous methods for determination of dissolution rate were based on recording the absorbance of the dissolution medium *versus* time. The percent of drug dissolved at any given time was obtained indirectly by comparing observed absorbance with absorbance of a solution representing total dissolution.

Use of the concentration converter eliminates this intermediate step.

EXPERIMENTAL

Because drug products are marketed in varying strengths, it was found most useful to calibrate the system in terms of a linear relationship between the drug concentration and the converter setting required for fullscale recorder deflection. The equation for this straight line is then used to calculate the setting required to display 0–100% dissolution for any label claim potency directly on the chart paper.

Six solutions of the drug of interest were prepared equivalent to 0–125% of label claim potency of the drug dissolved in a specified volume of dissolution medium.

A wavelength, preferably an absorption maximum, was selected for spectrophotometric monitoring. The zero concentration point was set with dissolution medium in reference and sample cells in the spectrophotometer. Each standard solution was then pumped through the sample cell, and the recorder deflection was adjusted to read 100 chart divisions by means of a calibrated potentiometer on the converter. The dial reading was interpolated to four places. A linear plot was obtained between the dial setting and concentration. The slope and y-intercept of the line were determined by the method of least squares. Abscissa values (x) for the equation were expressed

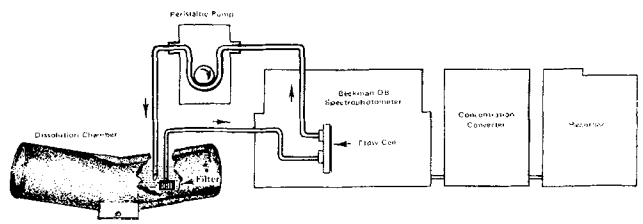

Figure 2—*Flow diagram of* in vitro *dissolution apparatus.*

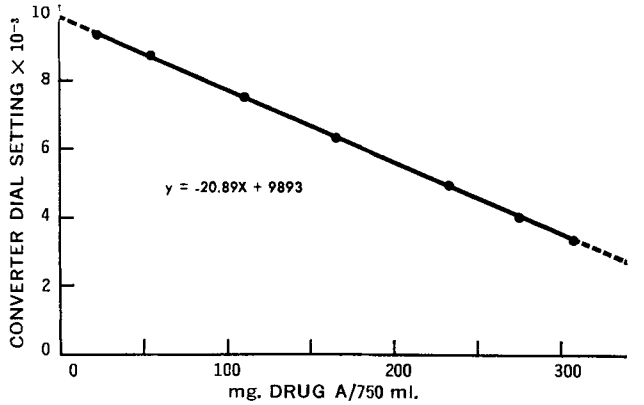

Figure 3—*Calibration curve for Drug A in simulated gastric solution (without pepsin) in 0.2-cm. flowcell at 364 nm.*

as milligrams of drug/volume of dissolution medium. The y-intercept for a given drug is constant regardless of the volume of test medium and y represents the calculated dial setting on the converter required for a given label claim potency in the required volume of dissolution medium. Slope depends on the volume of the dissolution medium and the lightpath of the cell. Thus, the straight-line equation for a particular tablet or capsule potency gives the dial setting required to display percentage dissolution *versus* time directly on the strip chart recorder over the range of 0–100% dissolution.

For example, 300-mg. film-coated tablets of Drug A were studied. Standard solutions were prepared with 0.1 N hydrochloric acid. A plot of concentration *versus* converter dial setting was linear, the least-squares fit yielding the equation $y = -20.89x + 9893$ at a wavelength of 364 nm. in a 0.2-cm. flowcell. The x is the label claim potency, and y is the calculated dial setting. For a label claim potency (x) of 300 mg., a dial setting (y) of 3626 is obtained.

At time zero, one tablet was placed into 750 ml. of simulated gastric solution USP (without pepsin) and agitated at a rate of 25 oscillations/min. This solution was pumped through the flowcell and returned to the chamber; drug concentration, in terms of percent dissolved, was recorded *versus* time.

RESULTS

Calibration plots for Drugs A, B, and C are shown in Figs. 3–5. The graphs were constructed from a least-squares fit of the experimental data. The strip chart recording of time *versus* percent dissolution for a 300-mg. film-coated tablet of Drug A is reproduced in Fig. 6. Prior to the use of a concentration converter, absorbance–time curves, such as that shown in Fig. 7, were obtained for similar tablets. In Fig. 7, the percent dissolved values are indicated at the appropriate absorbance values. One sees from Fig. 7 that percentage dissolution does not necessarily correspond logically to recorder deflection.

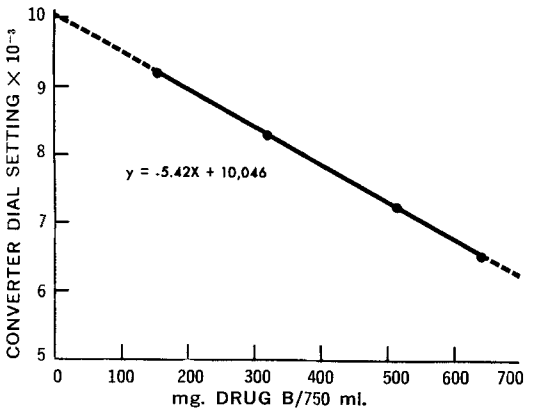

Figure 4—*Calibration curve for Drug B in simulated gastric solution (without pepsin) in 0.2-cm. flowcell at 276 nm.*

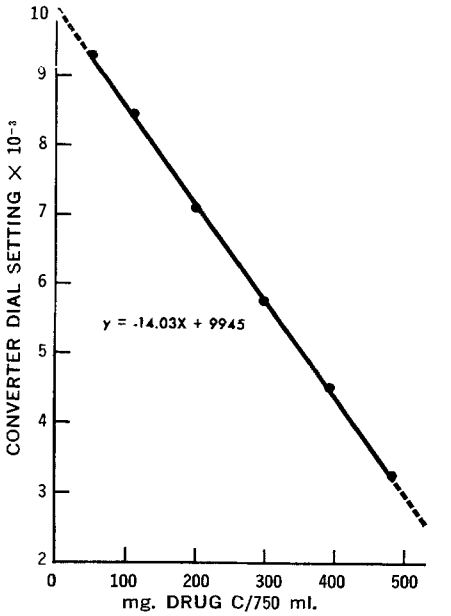

Figure 5—*Calibration curve for Drug C in simulated gastric solution (without pepsin) in 0.2-cm. flowcell at 380 nm.*

DISCUSSION

The calibration plots used here are based upon a specific volume of dissolution medium. It is easily recognized that a change in volume changes the slope of the least-squares line. Because of this, a set of least-squares coordinates and dial settings should be determined for each volume used. Slope also depends on absorptivity of the drug at the selected wavelength. Calibration curves for each of the three drugs studied had different slopes.

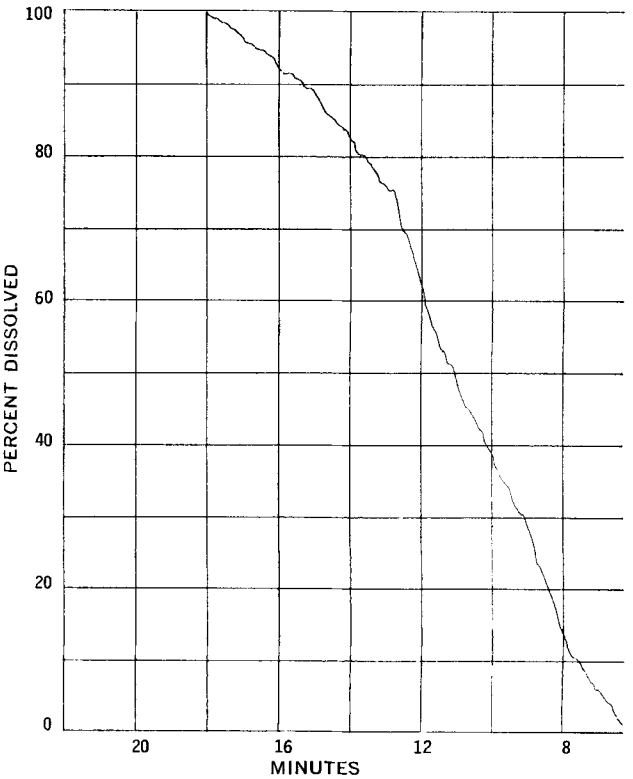

Figure 6—*Percent dissolved* versus *time: 300-mg. film-coated tablets of Drug A in simulated gastric solution (without pepsin); 0.2-cm. flowcell at 364 nm. Converter setting = 3526 for* $y = -20.89x + 9893$.

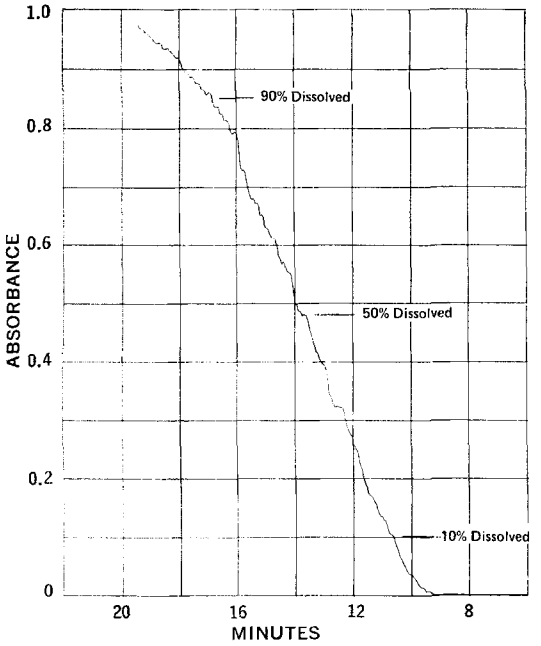

Figure 7—*Absorbance* versus *time: 300-mg. film-coated tablets of Drug A in simulated gastric solution (without pepsin); 0.2-cm. flow-cell at 391 nm.; 100% label claim concentration = 0.950 absorbance units.*

Values for *y*-intercepts appear to approach a limiting value of 9999+. This value is the highest number obtainable on the multipot dial. Although these numbers are arbitrary, 9999+ is analogous to 100% transmittance since the ordinate represents infinite dilution or 0% concentration.

In addition to monitoring concentration dynamics during a dissolution or kinetic study, the concentration converter allows for rapid analysis of solutions assayed by colorimetric or spectrophotometric methods. Samples can be quickly pumped through the flow-cell; their concentrations can be read directly from the chart rather than converting an absorbance value to concentration by means of absorptivity calculations or working curves.

While the primary advantage of the concentration converter in this study was direct display of a concentration parameter, the instrument also permits the presentation of absorbance and percent transmittance data on a recording chart simply by switching the multirange switch on the converter.

REFERENCES

(1) L. C. Schroeter, J. E. Tingstad, E. J. Knoechel, and J. G. Wagner, *J. Pharm. Sci.*, **51**, 865(1962).

(2) G. Levy, J. R. Leonards, and J. A. Procknal, *ibid.*, **54**, 1719 (1965).

(3) E. Nelson, *J. Amer. Pharm. Ass., Sci. Ed.*, **48**, 96(1959).

(4) J. A. Hersey, *Mfg. Chem. Aerosol News*, **Feb. 1969**, 32.

(5) H. Macdonald, F. Pisano, J. Burger, A. Dornbush, and E. Pelcak, *Clin. Med.*, **76** (12), 30(1969).

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NOTES

Absolute Configuration of (+)-*trans*-2-*o*-Tolyl-*trans*-5-hydroxycyclohexanol: Metabolite of Racemic *trans*-2-*o*-Tolylcyclohexanol

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Abstract □ The absolute configuration of the urinary metabolite (+)-*trans*-2-*o*-tolyl-*trans*-5-hydroxycyclohexanol, isolated after administration of racemic *trans*-2-*o*-tolylcyclohexanol in male Holtzman rats, was established by relating it chemically to (1S,2R)-(+)-*trans*-2-*o*-tolylcyclohexanol of known absolute configuration. The results give unequivocal proof that the original tentative assignment of (1S,2R,5R)-(+)-*trans*-2-*o*-tolyl-*trans*-5-hydroxycyclohexanol is correct.

Keyphrases □ (+)-*trans*-2-*o*-Tolyl-*trans*-5-hydroxycyclohexanol, urinary metabolite—absolute configuration □ Urinary metabolites—absolute configuration of (+)-*trans*-2-*o*-tolyl-*trans*-5-hydroxycyclohexanol

In an earlier publication (1), the authors reported the characterization of a major rat urinary metabolite of racemic *trans*-2-*o*-tolylcyclohexanol as the dextrorotatory axial C-5 ring hydroxylated product *trans*-2-*o*-tolyl-*trans*-5-hydroxycyclohexanol (IV). The characterization was done by NMR and by comparison with authentic racemic IV previously synthesized in this laboratory (2). In the same publication, a tentative assignment of absolute configuration of the metabolite was made by comparison of its ORD curve with that of (1S,2R)-(+)-*trans*-2-*o*-tolylcyclohexanol (V) (3). The assignment was tentative because of the lack of direct information on the effect of the remote hydroxyl group at C-5 on the Cotton effects of the aromatic chromophore at C-2.

Unequivocal proof is now presented that the original assignment of (1S,2R,5R) is correct. The proof of absolute configuration was obtained by relating (+)-*trans*-2-*o*-tolyl-*trans*-5-hydroxycyclohexanol (IV) chemically to (1S,2R)-(+)-*trans*-2-*o*-tolylcyclohexanol (V), the absolute configuration of which was previously reported (3) and recently confirmed by single crystal X-ray