Computerized Automated System for Determining Dissolution Rate Profiles for Solid Dosage Forms

F. J. CIOFFI ˣ, H. M. ABDOU, and A. T. WARREN

Abstract □ A unique automated system was developed for the measurement of dissolution rates of tablets and capsules. This system, which adheres to principles set forth in the compendia, can test six dosage forms simultaneously. It is modular in form and capable of maintaining sink conditions, and it can handle any type of chemistry amenable to an automatic analyzer. In addition to the usual dissolution apparatus, the system includes a 12-channel combination sampling and solvent addition pump, a sequence control module, pertinent automated analyzer components, a spectrophotometer or fluorometer, an optional recorder, a specially designed digitizing system, and a teletype equipped with a paper tape punch reader and acoustic coupler. Each dissolution flask is automatically sampled every 6 min. Standards may be run either before or after the samples. Since dissolution, sampling, chemistry, and readout are all accomplished simultaneously, the complete test time is essentially reduced to the dissolution time itself. At the completion of sample dissolution, raw dissolution profile data are on the punched paper tape ready for computer processing *via* a time-sharing system. Developed software provides for the printout of a complete test report in less than 15 min.

Keyphrases □ Dissolution—solid dosage forms, rate profiles determined using computerized automated system □ Computerized automated system—utilized for determining dissolution rate profiles of solid dosage forms □ Automated computer system—utilized for determining dissolution rate profiles of solid dosage forms □ Dosage forms, solid—tablets and capsules, dissolution rate profiles determined using computerized automated system

In recent years, the dissolution rates of solid dosage forms such as tablets and capsules have become of prime importance in pharmaceutical studies, as reflected by the inclusion in the compendia of monographs requiring the dissolution test. Once clinical correlations have been established, *in vitro* dissolution measurements can serve as a reliable, rapid, and economical technique to indicate the *in vivo* rate and extent of drug release and, hence, bioavailability (1–3). Dissolution testing also can serve as an important criterion in evaluating the effects of different constituents in drug formulations; in manufacturing, it can be an important method for determining uniformity both within and between different production batches.

BACKGROUND

Automation in dissolution rate testing has evolved in various ways and to different degrees. Three main approaches have been taken:

1. *Continuous flow* of dissolution fluid with the solid dosage form agitated by means of a motor-driven propeller, a rotating wire mesh basket containing the tablet[1], or some sort of rocking device (4–6).
2. Methods based on *mass transfer* between the solid dosage form and the dissolution fluid, with the tablet being held in a fixed position and traversed by a continuous flow of solvent. This concept may be referred to as the "flow-through" or "column" technique (7–9).
3. Methods based on *dialysis* (10).

In all three approaches, a constant volume is normally maintained in the dissolution flask throughout the test by techniques ranging from simply replacing manually the volume of the sample withdrawn by pure solvent to continuous dilution with pure solvent. In the former case, the dissolution curve is an integral function of the rate itself; the latter techniques can permit the maintenance of the sink condition, with the curve being a differential function of the rate.

Multiple automated dissolution testing systems were developed by a few investigators to relieve the ever increasing workload. Castello *et al.* (11) introduced an apparatus having multiple testing stations and capable of testing up to 20 tablets simultaneously. This apparatus, however, is really only a discrete sampling device, with the actual analysis being performed separately. Ferrari and Khoury (12) described a system capable of testing more than one tablet at a time. It uses a dialysis technique with the diffusate being fed into an automatic analyzer. This system, however, does not incorporate the official compendia dissolution apparatus, which has been generally accepted by the pharmaceutical industry.

Beyer and Smith (13) introduced a six-tablet apparatus in 1971 which does conform to compendia criteria. It consists of a dissolution apparatus in which seven flowcells[2] can be alternately positioned in the light beam of the spectrophotometer at programmed intervals. A single, seven-channel pump continually circulates the fluid from each of the six dissolution flasks through its respective cell and back again; the seventh cell is used for standardization with air. Absorbances are recorded on a strip-chart recorder in the form of bar graphs. Two limitations of this apparatus are that: (*a*) only direct spectrophotometric measurement of the "pure" dissolution fluid is possible, since the concept of this apparatus precludes sample dilution or reagent addition; and (*b*) sink conditions are not maintained.

The system developed in this laboratory also incorporates the USP–NF dissolution apparatus and can test six tablets simultaneously. Unlike other systems, however, it is completely automated from the point of tablet introduction to final readout of the raw data on punched paper tape. In addition, the system maintains sink conditions[3] at all times and performs any type of chemistry amenable to an automatic analyzer. The raw data are computer analyzed in minutes, and final results are presented in the form of a complete tabular report, a graphical dissolution profile, or both.

EXPERIMENTAL

Equipment—The system is modular, with the components being arranged as shown in Fig. 1. From left to right these components are:

Six-Spindle Dissolution Tester—It has a variable speed motor (25–250 rpm) and indicating tachometer[4]. This unit is shown in position over a 76.2 × 30.5 × 19.1-cm (30 × 12 × 7.5-in.) constant-temperature water bath[5] containing the six dissolution flasks[6]. The six shafts that hold the stainless steel wire mesh dissolution baskets[7] are shown extending from the chucks into the dissolution flasks.

12-Channel Peristaltic Pump[8]—The flow range is between 0.08 and 85 ml/min. Six channels pump sample continuously from each respective dissolution flask, while the other six channels return pure solvent to the dissolution flasks at the same flow rate. For standard measurements, only six channels are used. The six three-way stopcocks shown positioned above this pump allow solution to be sampled either from the dissolution flasks or from six standard containers.

Sequence Control Module (SCM)—This specially designed unit controls both the sampling sequence and the modified digital voltmeter (DDA) which, in turn, controls the electronic data acquisition

[1] From this point on, "tablet" will be used synonymously with "solid dosage form."

[2] Beckman Kintrac VII.

[3] Defined as never allowing the drug concentration in the dissolution flask to exceed 10–20% of saturation.

[4] Hansen model T-1044-20X, available from Scientific Glass Apparatus Co.

[5] Model 1064, Cole-Parmer.

[6] Corning No. 6947 1000-ml resin reaction flasks.

[7] Described in USP XVIII.

[8] Model 7555X Servodyne power drive system, Cole-Parmer, Chicago, Ill.

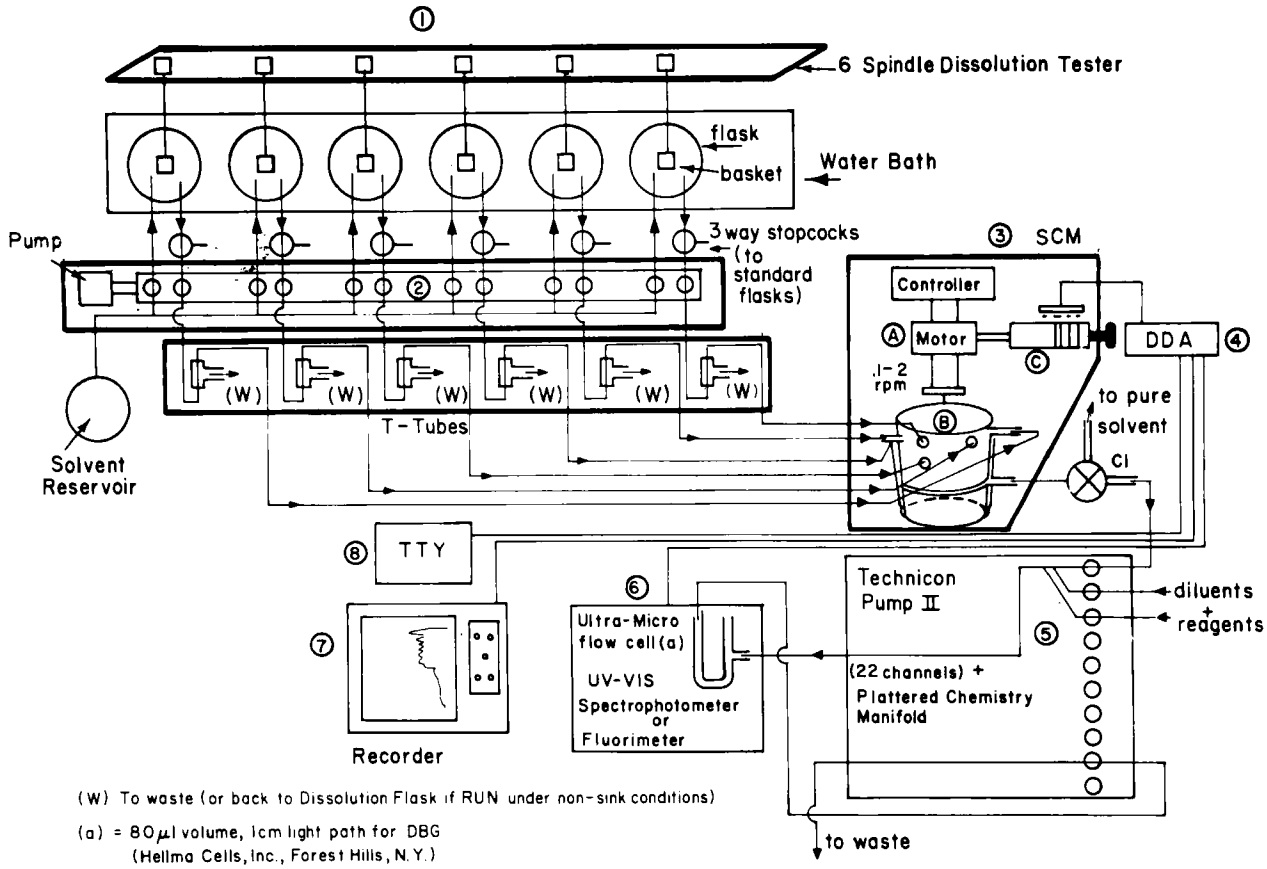

Figure 1—*Schematic of system (pat. 3,846,075).*

operations and the optional recorder. The sequence control module consists of a constant-speed 1/150 hp motor[9], which can be very accurately controlled over a range of 0–2 rpm by means of a controller[10] built into the front panel of the sequence control module. The double shaft of the motor is connected at one end to the sampling valve and at the other end, *via* a magnetic clutch[11], to a revolving plastic drum[12] having 60 equally spaced slots cut lengthwise along its outer cylindrical surface. Thus, sequence programming for the modified digital voltmeter unit is accomplished by placing small plastic actuators in the slots such that any of a number of microswitches may be contacted at desired intervals.

The sampling valve is a specially designed seven-port stopcock with a glass outer barrel and plug[13]. It is housed in the small compartment at the left side of the sequence control module. Figure 2 gives an operational diagram of the valve, showing the six equally spaced inlet ports (numbered 1–6), each connected to a separate dissolution flask *via* the sampling pump, and the single exit port (E). All glass ports attached to the outer barrel and the grooves shown on the plug are 1 mm i.d.

The length of the upper groove on the plug is 14/15 of the distance between the equally spaced inlet ports so that no two inlet ports can be opened at the same time as the plug rotates. The lower annular groove in the plug is connected to the upper groove by the vertical groove and is always open to the exit port. Thus, as the valve rotates, each dissolution flask is sampled in sequence, with the sample aliquot entering the automated chemistry system *via* the single exit port (E).

Digital Data Acquisition Module (DDA)—This modified unit[14] receives the raw analog data signal from the spectrophotometer and converts it to digital form for the teletypewriter printout while preserving the analog signal for input to the recorder, which it also controls. In addition, the digital data acquisition module controls the data output format on the teletypewriter. The "digitize period" wheel on the module sets the time in seconds over which the signal is averaged before it digitizes. This time should always be set to 4 sec or less to

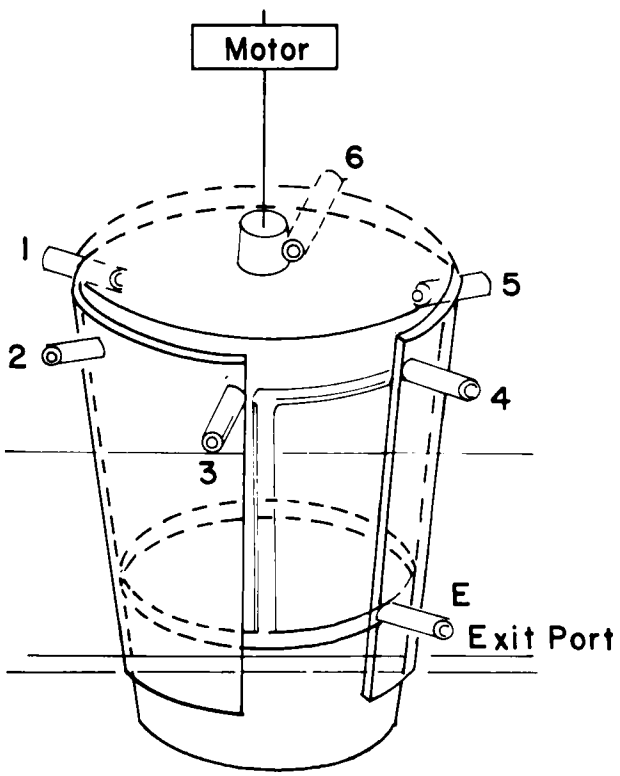

Figure 2—*Diagram of sampling valve (pat. 3,814,129).*

[9] Catalog No. B7096C-1080G, B&B Motor & Control Corp., New York, N.Y.
[10] Model MR-05, B&B Motor & Control Corp., New York, N.Y.
[11] Model SBEC clutch coupling, Electroid Co., Union, NJ 07083
[12] Model 091-1061-501 programming switch, Selectro Corp., Mamaroneck, NY 10543
[13] Teflon (du Pont).
[14] Infotronics model CRS-230 digitizer system.

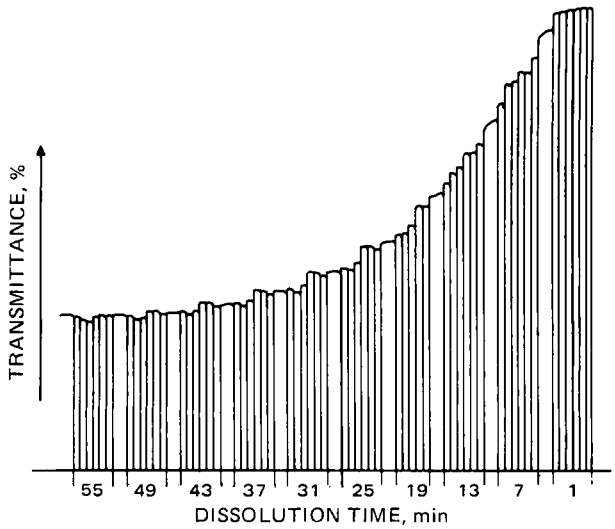

Figure 3—*Typical recorder chart.*

prevent any overlap between the signal averaging interval and the next digitize command.

The different control functions of this unit are activated by closure of four microswitches by the rotating drum of the sequence control module in a desired preprogrammed sequence. These functions are as follows:

1. Stand-By Mode—It resets the sample counter to 1 (Tablets 1–6 are numbered as such in the data printout).

2. Repetitive Mode—This microswitch returns the teletypewriter carriage, advances the line feed, and causes the next consecutive line number to printout.

3. Digitize Mode—The unit digitizes whenever this microswitch is closed.

4. Single Mode—Closure of this microswitch takes the unit out of the digitize mode, but manual digitization is still possible after this microswitch has been closed.

In addition to these functions, the digital data acquisition module also controls the recorder, which remains off most of the time. Before a run, the operator can set a selectable switch on the digital data acquisition module to any digitizing interval. The digital data acquisition module then automatically turns on the recorder only during this period for each tablet, the result being a series of bar graphs. Each set of six bar graphs corresponds to the degree of dissolution of Tablets 1–6, respectively, at the particular dissolution times noted and is separated by an elongated line for the sixth tablet value. An example of a typical recorder chart is shown as Fig. 3.

Since the sink condition is normally maintained by continuous dilution of the sample, as described earlier, the recorder bar graphs reflect only the actual concentration of drug in the dissolution flask at any time—not true amount dissolved. The recorder, therefore, usually serves only as an optional visual guide to the approach and maintenance of the sink condition. The same data, of course, are available in numerical form on the teletypewriter printout. However, with very soluble substances where it is not necessary to dilute the sample continually to maintain sink conditions, the system may be adjusted so that the recorder gives the actual percent dissolved directly.

The other modules shown in Fig. 1 include the proportioning pump (5), the spectrophotometer (6), the recorder (7), and the teletypewriter (8). Since the entire system is modular, it is quite simple to substitute different modules, *e.g.*, a fluorometer for the spectrophotometer.

The raw data output is obtained on the teletypewriter in both printed form and on punched paper tape. Each line represents a single tablet. Thus, under the usual preset conditions of 1-min sampling times for each flask (motor set to speed of ⅛ rpm) and a 6-sec digitizing interval program set on the drum, each tablet solution passes through the spectrophotometer flowcell for 1 min at 6-min intervals. Readings are taken automatically at 6-sec intervals for each tablet solution as it passes through the flowcell, beginning with 18 sec after the solution enters the cell. Since the standards are usually run at half this sampling rate to ensure steady state in the flowcell, readings are taken every 12 sec for each standard level, beginning 36 sec after the respective standard solution enters the cell.

Table I—Typical Standard and Sample Raw Data (on Punched Paper Tape)

				Standard Data				
	1	9802	9919	9992	9992	9992	6482	9448
	2	7879	7744	7621	7520	7390	7153	6848
	3	6024	5947	5913	5875	5816	5735	5649
	1	5383	5348	5332	5332	5344	5403	5522
	2	6126	6219	6298	6345	6387	6482	6731
	3	7823	7988	8123	8258	8411	5735	8815
1E25								
				Sample Data				
	1	4981	4975	5003	5002	4999	5003	5000
	2	4997	5000	5001	5005	5006	5002	5004
	3	4996	5001	5000	5009	5013	5005	5005
	4	4997	4998	5000	5002	5004	5009	5011
	5	5002	5000	5000	5000	5001	5005	5005
	6	5002	5006	5000	4998	5003	5001	5000
	1	5009	5027	5020	5021	5016	5012	5014
	2	5026	5023	5025	5021	5020	5015	5022
	3	5017	5018	5013	5015	5022	5019	5015
	4	5013	5007	5010	5008	5013	5012	5010
	5	5014	5012	5007	5019	5013	5006	5008
	6	5017	5015	5011	5016	5016	5016	5021
	1	6325	6283	6248	6210	6156	6107	6076
	2	6008	6010	6018	6020	6020	6034	6051
	3	6123	6133	6149	6178	6201	6217	6225
	4	6212	6188	6172	6175	6167	6152	6133
	5	6155	6177	6198	6250	6272	6279	6292
	6	6366	6371	6354	6352	6345	6335	6331
	1	6239	6199	6161	6122	6077	6046	6008
	2	5938	5941	5959	5982	6010	6020	6031
	3	6105	6120	6126	6151	6173	6188	6198
	4	6162	6147	6142	6052	6125	6126	6125
	5	6152	6150	6173	6186	6209	6221	6238
	6	6261	6271	6273	6259	6257	6254	6260
	1	6192	6153	6113	6072	6036	6004	5974
	1	5898	5903	5905	5917	5946	5975	6005
	2	6107	6125	6131	6116	6121	6133	6133
	3	6114	6098	6090	6082	6083	6095	6105
	4	6133	6133	6144	6149	6155	6160	6167
	5	6172	6167	6175	6174	6168	6167	6172
	1	6130	6093	6059	6019	5986	5940	5898
	1	5824	5829	5843	5868	5902	5920	5949
	2	6061	6082	6087	6093	6089	6078	6054
	3	6020	6022	6018	6024	6028	6043	6055
	4	6074	6078	6082	6087	6093	6090	6089
	5	6053	6045	6044	6043	6048	6056	6069
	1	6060	6019	5964	5929	5883	5841	5802
	2	5770	5782	5801	5838	5874	5913	5950
	3	6050	6042	6040	6037	6020	6017	6007
	4	5991	5969	5968	5977	5996	5993	6018
	5	6070	6069	6053	6050	6054	6057	6032
	6	5967	5964	5957	5957	5965	5968	5966
1E25								

Therefore, under the conditions specified, sample data printouts occur at 18, 24, 30, 36, 42, 48, and 54 sec after the sample enters the flowcell and standard data printouts occur at 36, 48, 60, 72, 84, 96, and 108 sec after the standard solutions enter the cell. An example of typical raw data output is shown as Table I. The columns of standard and sample data that most closely represent steady-state readings are easily noted by visual inspection. These columns (usually numbers 4 or 5 for the standards and 5 or 6 for the samples) are used in the computer calculations.

Operation—After setting up the appropriate automated chemistry platter on the proportioning pump[15], the teletype is switched to "DATA SYSTEM." The procedure for starting the system running automatically is as follows.

Standards

1. Turn stopcock C1 (Fig. 1) to the solvent position. This three-way stopcock is positioned to take solution from the sampling valve exit port during the sample run and standard measurements. It is positioned toward a flask containing pure solvent when rapid flushing of the system is desired to obtain a pure solvent baseline reading.

2. The six three-way stopcocks attached to the six alternate sampling channels of the 12-channel pump are positioned toward the six flasks containing the standard solutions; the remaining six solvent return channels are not used.

[15] Technicon.

AUTOMATED DISSOLUTION TEST

Product: An Antibiotic Tablet Formulation
Code: None
Lot Number: Red Oval
Control: KDN-140-UJB1

Date Tested: 3-15-73
Analyst: HA

Solvent: Simulated Intestinal Fluid W.O. Pancreatin
Volume: 1000ml
Basket Rotation Rate: 150RPM

F = 19.3 ml/minute
T = 37 deg.c

Percent Dissolved (Corrected to Sink Conditions)
(Basis-Label)

Time (Min)	Tablet Number 1	2	3	4	5	6	Min.	Max.	Avg.
1	3.46	4.72	4.52	5.27	5.40	9.06	3.46	9.06	5.41
7	10.46	10.88	11.66	12.96	15.44	21.84	10.46	21.84	13.88
13	22.13	23.11	24.29	24.21	27.33	31.66	22.13	31.66	25.45
19	32.55	33.39	33.67	35.58	37.33	41.77	32.55	41.77	35.71
25	41.21	41.12	43.27	44.59	47.61	48.33	41.12	48.33	44.35
31	48.34	48.92	52.01	53.87	54.25	53.46	48.34	54.25	51.81
37	53.29	52.87	56.58	58.37	60.43	57.62	52.87	60.43	56.53
43	55.72	57.33	58.97	61.53	64.63	61.10	55.72	64.63	59.88
49	58.86	60.71	62.46	66.42	67.77	63.48	58.86	67.77	63.28
55	62.55	63.15	66.71	68.10	69.18	65.22	62.55	69.18	65.82
61	64.21	64.26	69.00	70.95	72.15	68.06	64.21	72.15	68.10
67	66.66	66.87	70.95	73.83	75.47	70.74	66.66	75.47	70.75
73	69.14	69.89	74.63	76.84	76.87	74.31	69.14	76.87	73.61
79	73.63	73.10	79.03	79.97	79.42	77.15	73.10	79.97	77.05
85	77.55	76.37	82.51	83.92	83.40	80.10	76.37	83.92	80.64
91	81.11	80.70	86.91	86.64	86.05	83.43	80.70	86.91	84.14
97	84.07	83.19	90.09	89.55	88.05	86.08	83.19	90.09	86.84
103	86.27	86.19	92.54	91.67	91.87	87.98	86.19	92.54	89.42
109	89.76	89.13	95.26	96.07	93.09	90.80	89.13	95.26	92.08
115	90.79	90.12	96.45	96.83	95.67	92.59	90.12	96.83	93.74
121	93.09	92.16	100.33	99.37	98.20	93.77	92.15	100.33	96.15
127	96.02	95.43	103.64	102.54	101.17	98.07	95.43	103.64	99.48
133	97.45	96.46	106.37	103.69	103.34	98.73	96.46	106.37	101.01

DT (50) = 29.5 minutes
DT (85) = 92.9 minutes

Figure 4—*Typical computer printout of final dissolution test report.*

3. The 12-channel pump is then set to a relatively fast speed for about 2 min to make sure that the six tubes leading to the sampling valve are filled with their standard solutions. Then, by using a preconstructed calibration curve, the speed is reduced to about twice the sampling speed of the proportioning pump; the excess standard solution exits *via* the overflow T-tubes either to waste or back to its standard flask.

4. The program drum is set manually by means of a position-indicating wheel attached to the drum through the sequence control module case such that the "STAND-BY" microswitch is just closed as indicated by the "STAND-BY" light on the digital data acquisition module. The magnetic clutch is not yet engaged.

5. The sequence control module motor (A in Fig. 1) speed is set to 12 min/revolution, equivalent to a 2-min sampling time for each standard solution.

6. When the buzzer sounds, indicating that the sampling valve has just reached the Sample 1 position, stopcock C1 (Fig. 1) is turned to sample from the exit port of the sampling valve and a stopwatch is started. Standard 1 is now being drawn into the automated chemistry system, and all six standard levels will be sampled automatically, sequentially, and repetitively for 2-min intervals. The relative concentration of the standard levels decreases from 1 to 6, with the range in terms of percent of label being from about 110 to 20% of the maximum concentration of drug in the dissolution flask at the sink condition maintained. This reduced absolute concentration range is used to improve the linearity of the standard curve.

To minimize any cumulative effect of carryover in the flowcell, the sampling order used for the standards is 1, 3, 5, 6, 4, 2. When the first (and most concentrated) standard reaches the flowcell, as indicated by the first deflection of the spectrophotometer needle, the stopwatch is stopped and the switch engaging the programming drum is thrown. The time for the solution to move from the sampling valve to the flowcell is noted as $T1$. As the standards are automatically run, the raw data (transmittance or absorbance) are recorded on the teletype and on punched paper tape.

Samples—Except for a few obvious differences, the procedure for

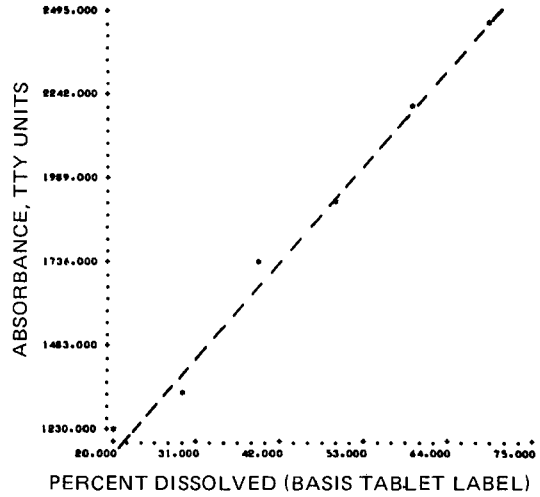

Figure 5—*Computer-drawn standard curve.*

running the samples is similar to that for the standards.

1. All channels of the 12-channel peristaltic pump are used. The six channels not used for the standard run are used to add pure solvent to the tablet dissolution flasks.

2. The six baskets containing the test tablets are positioned above their respective dissolution flasks. At exactly $(6 - x)$ min[16] from the first sound of the buzzer after a satisfactory baseline has been obtained, the basket containing Tablet 1 is lowered into its dissolution flask and rotation is begun. Tablets 2–6 are then started at 1-min intervals.

3. At exactly $(T1 + x)$ min after Tablet 1 was started, the program drum is engaged and the tape punch is turned on. At this point, Sample 1 is just entering the flowcell. The run now proceeds automatically until complete dissolution is obtained and the system is manually stopped.

Data Handling—The data tape is fed into a time-share computer system, and all calculations are done *via* software developed in this laboratory. The standard data are fit to six possible equations, and the equation that best fits the data is chosen by the computer for further calculations. The percent dissolved (uncorrected for dilution) is then calculated for each tablet at 6-min intervals up to complete dissolution. In addition, the high, low, and average results of the six tablets are also calculated for each time interval. At this point, the results are corrected for dilution based on the method of Richter *et al.* (6) as follows.

The rate of elimination of dissolved drug under constant volume conditions may be expressed as:

$$E = F/V \qquad \text{(Eq. 1)}$$

where F = flow rate in milliliters per minute, and V = constant volume in dissolution flask in milliliters.

The rate of change of concentration in the solution (neglecting continued dissolution of undissolved drug) can be expressed as:

$$-\frac{dc}{dt} = (E)(c) \qquad \text{(Eq. 2)}$$

where c = concentration in milligrams per milliliter.

Integrating between the limits c_0 and c_t gives:

$$\int_{c_0}^{c_t} \frac{dc}{c} = -E \int_0^t dt \qquad \text{(Eq. 3)}$$

or:

$$c_t = c_0 e^{-Et} \qquad \text{(Eq. 4)}$$

where c_t = concentration of dissolved drug at any time, t, in milligrams per milliliter, and c_0 = concentration of dissolved drug at time zero. Thus, the rate of change of the drug concentration in solution under constant volume conditions is first-order kinetic.

As the tablet dissolves, the concentration of drug in the dissolution flask builds up to a peak value after which, because of continuous dilution, it begins to decrease. Thus, before the peak, the amount of drug dissolved may be calculated as follows. During the first interval, Δt, the amount of drug released is given by:

$$C_1 = c_1 + c_t \qquad \text{(Eq. 5)}$$

where c_1 = amount of drug *observed* in solution, and $c_t = c_0 e^{-Et}$, the amount of drug *eliminated* from solution (Eq. 4).

If the sampling time interval is small[17], then Eq. 4 may be simplified to:

$$c_t = c_1 \, \Delta t E \qquad \text{(Eq. 6)}$$

and Eq. 5 becomes:

$$C_1 = c_1 + c_1 \, \Delta t E \qquad \text{(Eq. 7)}$$

For the next interval, Δt, the amount of drug released is calculated in a similar manner:

$$C_2 = c_2 + c_1 \, \Delta t E + c_2 \, \Delta t E = c_2 + \Delta t E(c_1 + c_2) \qquad \text{(Eq. 8)}$$

This calculation is done for every time interval up to the peak in the curve, where the amount of drug released is given by:

$$C_p = c_p + E \, \Delta t(c_1 + c_2 \ldots c_p) \qquad \text{(Eq. 9)}$$

or:

$$C_p = c_p + E \, \Delta t \sum_{t=1}^{t=p} c_t \qquad \text{(Eq. 10)}$$

After the peak, if there is no further release of drug from the solid dosage form, the elimination of the dissolved drug will follow Eq. 4. However, if the amount of drug observed in solution after the next time interval is higher than that calculated by Eq. 4, then the solid dosage form is still dissolving and releasing drug into solution. Hence, during the first time interval after the peak, the amount of drug released is given by:

$$C_{p+1} = c_{p+1} - c_p e^{-E \, \Delta t} \qquad \text{(Eq. 11)}$$

where C_{p+1} = amount of drug released during the first time interval after the peak, c_{p+1} = amount of drug observed in solution, and $c_p e^{-E \, \Delta t}$ = amount of drug expected in solution due to pure dilution after the first interval (by Eq. 4).

For the second time interval after the peak, the amount of drug released from the solid dosage form will be:

$$C_{p+2} = c_{p+2} - (c_{p+1})e^{-E \, \Delta t} \qquad \text{(Eq. 12)}$$

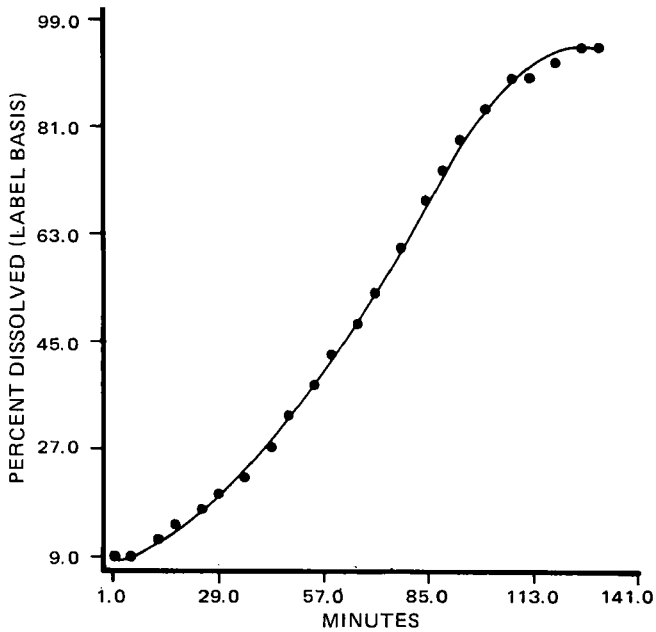

Figure 6—*Computer-drawn dissolution profile curve.*

[16] The x is the time required for the solution to be pumped from the dissolution flasks to the sampling valve at the particular flow rate being used. Since the length of tubing is the same from each flask, x is the same from each flask and is known from a calibration table constructed beforehand for any flow rate setting of the peristaltic pump.

[17] Satisfactory results were obtained for sampling intervals of up to 10 min.

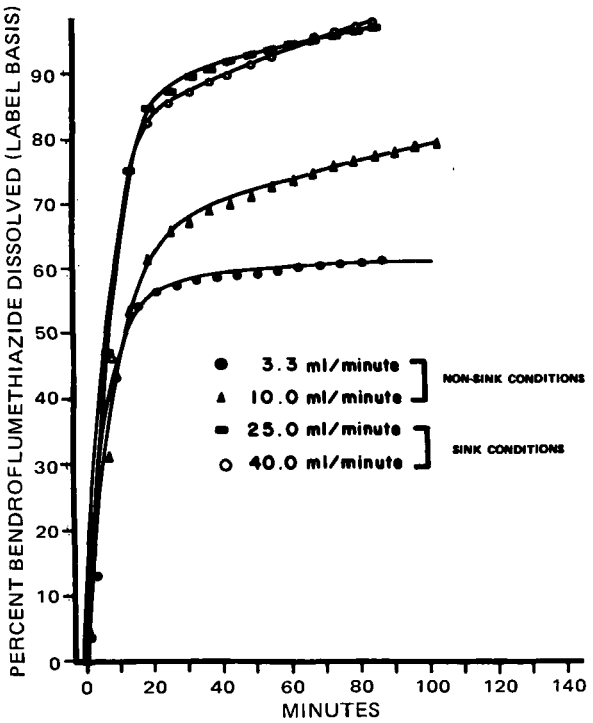

Figure 7—*Effect of flow rate on approach to sink conditions.*

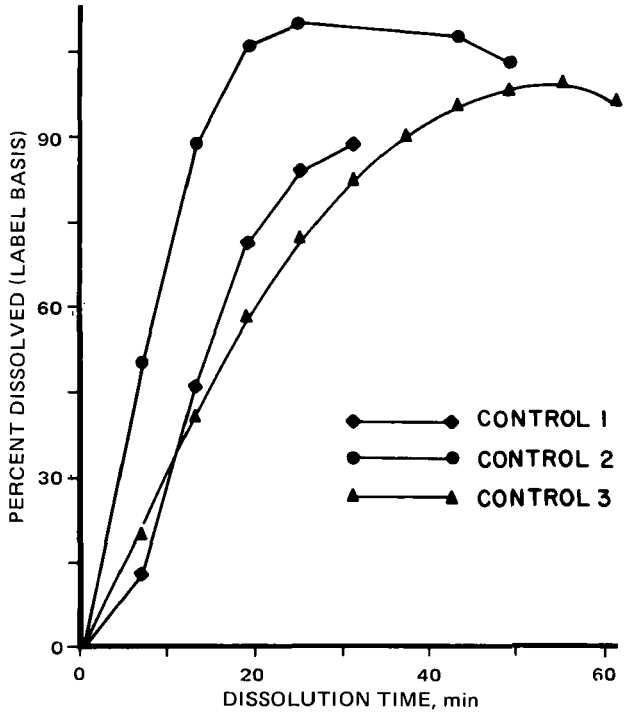

Figure 8—*Computer-drawn series of dissolution profiles.*

In general form, then:

$$C_{p+n} = c_{p+n} - (c_{p+(n-1)})e^{-E\,\Delta t} \qquad \text{(Eq. 13)}$$

where C_{p+n} = amount of drug released during any time interval, n; c_{p+n} = amount of drug observed in solution after n − time intervals after the peak; and $(c_{p+(n-1)})e^{-E\,\Delta t}$ = amount of drug expected in solution after $(n - 1)$ time intervals after the peak (by Eq. 4). When:

$$c_{p+n} = (c_{p+(n-1)})e^{-E\,\Delta t} \qquad \text{(Eq. 14)}$$

Eq. 13 becomes:

$$C_{p+n} = 0 \qquad \text{(Eq. 15)}$$

and there is no further release of drug. Therefore, the total amount of drug released (or dissolved) from the solid dosage form is obtained by combining Eqs. 10 and 13, *i.e.*:

$$C_T = C_p + \sum_{q=p+1}^{q=p+n} C_q \qquad \text{(Eq. 16)}$$

where C_T = total amount of drug released (dissolved from solid dosage form), C_p = amount of drug released before peak, and C_q = amount of drug released after peak. After the dilution correction is applied, the final results are presented in a tabular report format.

RESULTS AND DISCUSSION

An example of a typical computer-generated dissolution test report is shown in Fig. 4[18]. In this case, the product was an antibiotic tablet formulation. Figure 5 shows an optional printout of the standard curve, and Fig. 6 shows an optional printout of the dissolution profile curve for these tablets.

The importance of maintaining sink conditions by using an ample flow rate is clearly demonstrated in Fig. 7, which shows the effect of the flow rate on the dissolution profiles of the very insoluble bendroflumethiazide in a tablet formulation. NF XIV gives the solubility as "practically insoluble in water" (1 part in > 10,000 parts of water). Laboratory measurement showed a solubility of only 37.3 mg of bendroflumethiazide/liter of 0.1 *N* HCl, the dissolution fluid used. As Fig. 7 shows, a flow rate of 25 ml/min was necessary to maintain the calculated sink condition of +3.7–7.5 mg/ml in the dissolution flask.

Figure 8 shows an example of a computer-drawn set of dissolution profiles for three controls of the same product. In this case, the profile data for each control are stored in the computer as they are calculated and later combined into the single graph shown by using a commercial[19] package. Graphs such as these provide rapid visual observation of differences in the dissolution characteristics between different batches and thus provide an excellent technique for monitoring uniformity in product manufacture.

In addition to the drugs mentioned in the figures, the system described has been used to determine the dissolution profiles of more than 70 other tablet and capsule formulations. These include various antibiotics, steroids, vitamins, sedatives, diuretics, antituberculins, and antifilarial drugs and utilize UV, colorimetric, and fluorescence assays. Some results will be reported in forthcoming articles, along with *in vitro–in vivo* comparison studies.

CONCLUSION

A computerized automated dissolution testing system has been described which conforms to the basic requirements of the USP and NF dissolution test. The system, which can accommodate almost any type of tablet agitation technique, can test six dosage forms simultaneously under sink or nonsink conditions and can test products requiring any type of chemistry amenable to an automatic analyzer. The total run time is reduced essentially to the sample dissolution time. Finally, a complete dissolution profile report, with or without graphs, is obtained within minutes after the required sample dissolution time.

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Sensitive GLC Procedure for Simultaneous Determination of Phenytoin and Its Major Metabolite from Plasma following Single Doses of Phenytoin

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Abstract □ An improved GLC procedure was developed for the simultaneous determination of phenytoin and its metabolite, 5-(*p*-hydroxyphenyl)-5-phenylhydantoin, in plasma and urine following enzyme hydrolysis. After extraction, the drug, the metabolite, and the internal standard, 5-(*p*-methylphenyl)-5-phenylhydantoin, are measured by GLC with flame-ionization detection as their respective methyl derivatives following flash-heater methylation with trimethylanilinium hydroxide. The drug and metabolite give well-resolved symmetrical peaks on a phenyl methyl silicone column, and the method has a sensitivity of 150 ng/ml of phenytoin and 125 ng/ml of the metabolite. GLC–mass spectral evidence is presented for the formation and intact determination of methyl derivatives of the drug, its metabolite, and the internal standard.

Keyphrases □ Phenytoin and major metabolite—simultaneous GLC analysis, plasma □ GLC—simultaneous analysis, phenytoin and major metabolite, plasma □ Anticonvulsant agents—phenytoin, GLC analysis, plasma

Phenytoin is a commonly prescribed anticonvulsant drug. The various procedures used for drug level measurements were reviewed (1). It is one of the few drugs for which plasma concentrations are routinely monitored, and many GLC methods (2–14) are suitable for the assay of steady-state levels. The dose-dependent biotransformation of phenytoin and variations in its metabolism in treated subjects require that assay methods for determining plasma concentrations of the unchanged drug and its principal metabolite, 5-(*p*-hydroxyphenyl)-5-phenylhydantoin (15), suitable for single-dose pharmacokinetic studies be developed.

Chang and Glazko (8) reported the preparation of trimethylsilyl derivatives applied to the GLC assay of phenytoin in plasma and 5-(*p*-hydroxyphenyl)-5-phenylhydantoin in urine. However, when this procedure is used routinely, traces of moisture cause problems and the sensitivity is limited. Derivatization with diazomethane was applied in procedures based on GLC estimation of 3-*N*-methyl derivatives (2, 4).

The safer tetramethylammonium hydroxide reagent also was used in a flash-heater methylation procedure (6), while trimethylanilinium hydroxide was described as a methylating agent for analysis of phenytoin and its metabolite (13). However, in the assay procedure for the latter, acid hydrolysis was used to degrade the conjugate and the method required two internal standards. Although the unconjugated metabolite could be measured (13), the plasma levels were not given for the metabolite and no variation or sensitivity limit was reported.

Recently, tetramethylammonium hydroxide was used for methylating phenytoin and its metabolite along with subsequent quantitative estimation by GLC with temperature programming (14). This reported procedure is cumbersome and is only applicable when plasma concentrations of phenytoin and the metabolite are around 1 μg or more. In this report, a modified method is described for the GLC analysis of plasma phenytoin and total 5-(*p*-hydroxyphenyl)-5-phenylhydantoin by formation of methyl derivatives with trimethylanilinium hydroxide. The procedure is of sufficient sensitivity to allow the quantitation of 0.15 μg/ml of phenytoin and 0.125 μg/ml of the metabolite. In addition, GLC–mass spectral evidence is presented for the formation and determination of methyl derivatives of the drug and the metabolite used in the quantitation.

EXPERIMENTAL

Reagents—Ether[1] was glass distilled prior to use. Stock solutions containing 100 μg/ml of phenytoin were prepared by dissolving appropriate amounts of the sodium salt of phenytoin[2] in distilled water. Stock solutions containing 100 μg/ml of 5-(*p*-hydroxyphenyl)-5-phenylhydantoin[3] were prepared by dissolving the appropriate amount in 0.01 *N* NaOH. Appropriate dilutions of the drug (0.15–8.0 μg/ml) and the metabolite (0.125–4.0 μg/ml) were prepared as required.

Aqueous solutions (100 μg/ml) of the internal standard, 5-(*p*-methylphenyl)-5-phenylhydantoin[3], were prepared by dissolving an appropriate amount of the internal standard in 0.01 *N* NaOH and

[1] Ether (anhydrous), Mallinckrodt Chemical Works Ltd., Montreal, Canada.
[2] Diphenylhydantoin-Na, Parke-Davis & Co., Brockville, Ontario, Canada.
[3] Aldrich Chemical Co., Milwaukee, Wis.