(1980).
(18) N. Shane and R. Stillman, *ibid.*, **60,** 114 (1971).
(19) H. Bundgaard and C. Bundgaard, *J. Pharm. Pharmacol.*, **25,** 593 (1973).
(20) R. D. Kirchhoefer and W. E. Juhl, *J. Pharm. Sci.*, **69,** 548 (1980).
(21) R. D. Kirchhoefer, J. C. Reepmeyer, and W. E. Juhl, *ibid.*, **69,** 550 (1980).
(22) E. G. C. Clarke, "Isolation and Identification of Drugs," W. Clowes and Sons, London, England, 1969, p. 538.
(23) I. Sunshine and S. R. Gerber, "Spectrophotometric Analysis of Drugs," Charles C Thomas, Springfield, Ill., 1963, p. 27.
(24) R. J. Henry, D. C. Cannon, and J. W. Winkleman, "Clinical Chemistry–Principles & Technics," Harper and Row, New York, N.Y., 1974, p. 1592.
(25) A. Weller, *Z. Elektrochem.*, **60,** 1144 (1956).
(26) S. G. Schulman, P. J. Kovi, and J. F. Young, *J. Pharm. Sci.*, **62,** 1197 (1973).
(27) W. L. Paul and S. G. Schulman, *Anal. Chem.*, **45,** 415 (1973).
(28) J. F. Ireland and P. A. H. Wyatt, "Advances in Physical Organic Chemistry," vol. 12, Academic, New York, N.Y., 1976, pp. 193–195.
(29) I. Sunshine, "Methodology for Analytical Toxicology," CRC Press, Cleveland, Ohio, 1975, pp. 342, 343.

ACKNOWLEDGMENTS

The authors thank the Wayne State University and the Wayne County Medical Examiner's Office for the use of their facilities.

Continuous Flow Bead-Bed Dissolution Apparatus for Suppositories

T. J. ROSEMAN ˣ, G. R. DERR, K. G. NELSON, B. L. LIEBERMAN, and S. S. BUTLER

Received August 1, 1980, from the *Pharmacy Research Unit, The Upjohn Company, Kalamazoo, MI 49001.* Accepted for publication December 2, 1980.

Abstract □ A bead-bed dissolution apparatus for suppositories was evaluated by measuring the release of benzocaine from various vehicles. During dissolution, suppositories soften, deform, disintegrate, and eventually pass through a phase change from a solid to an oil. The control of the interfacial area during dissolution is a key factor in obtaining experimentally reproducible release data. The proposed suppository dissolution apparatus was designed to provide greater constancy of the exposed suppository area for dissolution. The apparatus consisted of a glass bead-bed containing the suppository. A continuous flow of liquid was passed through the bead-bed at a constant rate. Direct contact of the suppository was maintained with the dissolution medium, confining the suppository within the beads.

Keyphrases □ Suppositories—evaluation of a bead-bed dissolution apparatus, drug release rate □ Dissolution—bead-bed apparatus for suppositories, evaluation, drug release rate □ Drug release—suppositories, evaluation of a bead-bed dissolution apparatus

Benzocaine was selected as a model compound with both a low melting (33.5–35.5°) and a high melting (37–39°) glyceride-type suppository base. *In vitro* release of benzocaine decreased as the melting point of the suppository increased. Reproducibility of the complete release curves was acceptable. Drug release also was affected by the temperature of the dissolution media, increasing, decreasing, and increasing again at certain temperatures. This finding was related to the ability of the beads to penetrate the surface of the suppository when the suppository softened. Release profiles, however, were reproducible at the temperature studied. The proposed bead-bed dissolution apparatus should offer an improved means for measuring drug release from suppositories.

BACKGROUND

Suppositories, administered either vaginally or rectally, are utilized as a dosage form for various drugs (1). Recent reports investigated the use of vaginally administered prostaglandins for fertility control and also emphasized certain advantages of this mode of drug therapy, *e.g.*, self-administration and single-dose therapy (2–9). This continued interest in suppositories and suppository bases (10) has led to the recognition that a dissolution test would be helpful during the initial phase of dosage form design. In addition, such a test would provide valuable information on the effect of storage time and temperature on the subsequent *in vitro* release profile.

The methods used for testing drug release rate characteristics of suppositories *in vitro* can be classified in terms of five general types (Fig. 1). The first type consists of simple placement of the suppository in a flask or beaker (11–15). The second type utilizes an existing tablet dissolution apparatus that provides a wire mesh basket for holding the sample (16–22). The third and fourth types employ a membrane; the third consists of a sample chamber separated from a reservoir by a membrane (23–33), whereas the fourth employs dialysis tubing or a natural membrane (33–46). The fifth type involves a flow system in which the sample is placed on cotton or a wire screen (47, 48).

One basic problem in testing for drug release from a suppository is that the suppository softens, deforms, melts, or disintegrates during the test, exposing a variable interfacial area to the dissolution medium. Because the release rate is dependent on the interfacial area, the variability of this factor leads to poor test reproducibility.

Membranes have been used to control the interfacial area on the principle that when the suppository softens, it would spread over the entire membrane, restricting the area exposed to the dissolution fluid. Bhavnagri and Speiser (24) designed a type three apparatus with a relatively small sample chamber. Others (39, 42) used a relatively small bag in a type four apparatus to restrict the interfacial area.

The need to control the interfacial area is important, but the intro-

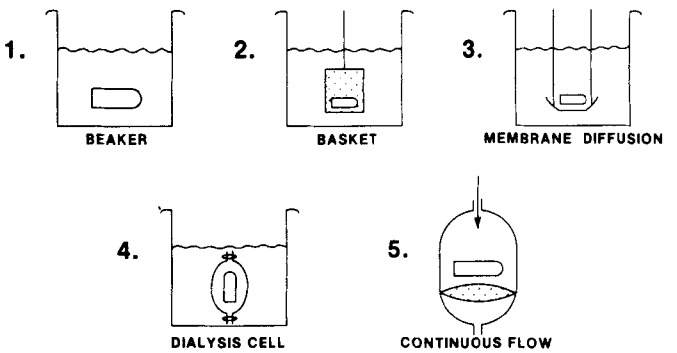


Figure 1—*Suppository dissolution methods.*

0022-3549/81/0600-0646$01.00/0

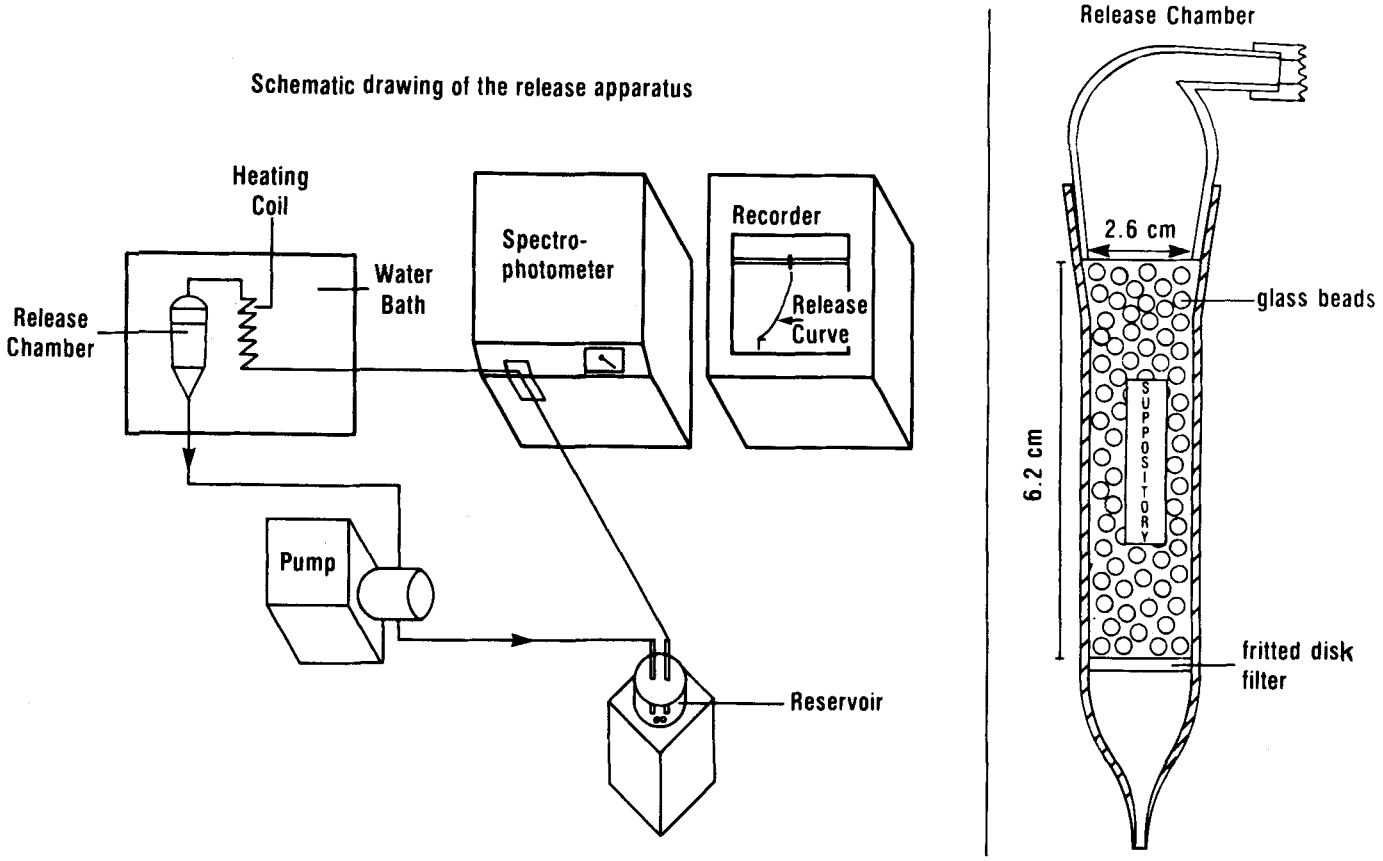


Figure 2—*Continuous flow bead-bed suppository dissolution apparatus.*

duction of an additional physical process, *i.e.*, membrane transport, complicates matters and may mask the real release characteristics for certain drug–suppository base combinations. Therefore, a flow-through technique was developed in which the suppository is secured in a bed of glass beads. This arrangement controls the interfacial area for most suppository bases, yet allows direct contact between the suppository and the dissolution medium.

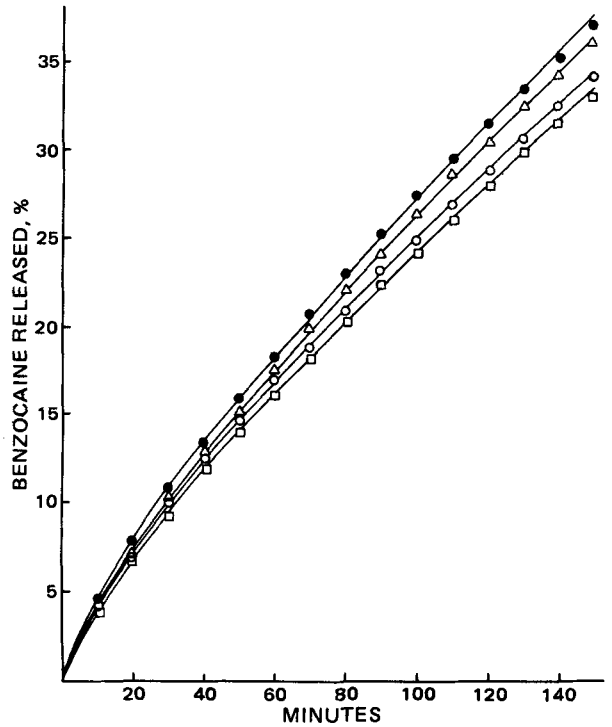


Figure 3—*Release of benzocaine from Base A suppositories on different days at 37° (original dissolution chamber). Key: ●, Day 1; ○, Day 2; □, Day 3; and △, Day 3.*

EXPERIMENTAL

Suppository Preparation—The suppositories were prepared by first dissolving an appropriate amount of benzocaine[1] (ethyl *p*-aminobenzoate) in the melted base to yield a benzocaine concentration of 0.125% (w/w). The suppository bases, designated A[2] for the low melting (33.5–35.5°) and B[3] for the higher melting (37–39°), were melted at 5–10° above their melting points. The melt then was poured into aluminum suppository molds and allowed to cool at room temperature, and the excess base was scraped away after solidification. The molds were cooled further at room temperature (Base A) or in a refrigerator (Base B) for 30 min, and then the suppositories were ejected from the molds.

The molds consisted of tapered cavities drilled into an aluminum block. The taper permitted easy removal of the suppositories. The suppositories were 800 mg, 1.91 cm long, and 0.75 and 0.79 cm in diameter at the ends. The amount of benzocaine in each suppository was 1.0 mg.

Design of Release Apparatus—The release rate apparatus was designed to provide reasonable control over the interfacial area for drug dissolution (Fig. 2). A continuous eluent flow is maintained over the suppository contained in a bed of glass beads. The eluent starts in the 150-ml jacketed beaker, which is covered with an acrylic resin disk. When the pump[4] is started, water is drawn through polyethylene tubing (3 mm o.d. × 2 mm i.d.) from the 150-ml jacketed beaker to a 1-cm flowcell in the UV spectrophotometer[5]. From the spectrophotometer, the water is drawn into a preheating glass coil (glass automated analyzer coil, 1.5 cm in diameter × 16 cm in length). The glass coil is submerged in the water bath[6] to allow the water temperature to equilibrate before reaching the sample chamber, which also is submerged in the water bath. The water leaves the glass coil and enters the sample chamber from the top.

[1] Pfaltz & Bauer, Stamford, Conn.
[2] Witepsol H-15, Dynamit Nobel Chemical, Oberlar, West Germany.
[3] Witepsol E-76, Dynamit Nobel Chemical, Oberlar, West Germany.
[4] Lab pump RRP, Fluid Metering Inc., Oyster Bay, N.Y.
[5] Beckman DB with recorder, Beckman Instruments, Fullerton, Calif.
[6] Tamson PMT, Neslab Instruments, Portsmouth, N.H.

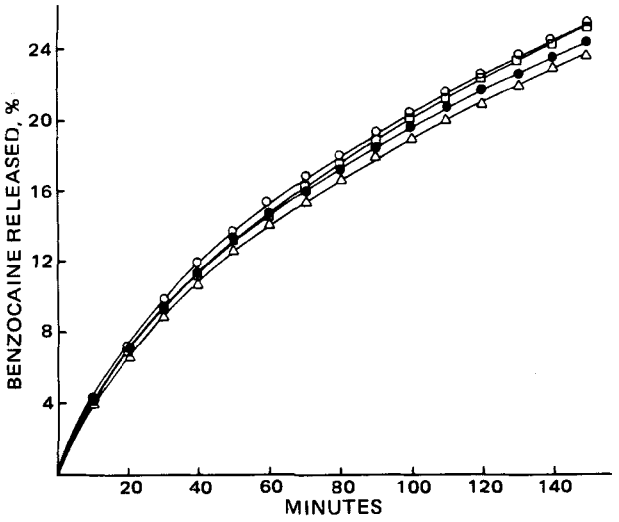


Figure 4—*Release of benzocaine from Base B suppositories on different days at 37° (original dissolution chamber). Key: ●, Day 1; ○, Day 1; □, Day 3; and △, Day 3.*

Originally, the chamber consisted of two 15-ml Büchner fritted disk filters connected at the top by Tygon tubing and a clamp. The present chamber consists of two sections. The bottom section is a 15-ml coarse Büchner fritted disk filter modified with a 24/40 glass joint at the top and an outlet tube narrowed to 4 mm o.d. × 2 mm i.d. The top half is a 24/40 hose-connecting joint modified with an inlet tube narrowed to 4 mm o.d. × 2 mm i.d. The water travels through the glass bead-bed (3-mm diameter, chemically resistant glass balls) and around the suppository. The dimensions of the cell containing the beads is 2.6 cm i.d. and 6.2 cm long. The original chamber was 2.3 cm i.d. and 4.6 cm long. The water leaves the test chamber, is drawn into the pump, and is pumped back into the 150-ml jacketed beaker, which is stirred by a magnetic stirrer.

Procedure—Before a release rate experiment was begun, a water baseline was established by adding 130 ml of deionized water to the 150-ml jacketed beaker. The water bath was adjusted to the appropriate temperature, and the system and water were allowed to equilibrate. Temperature was controlled within ±0.1°. The test chamber then was half-filled with glass beads. A weighed suppository was inserted into the center of the chamber, using a plunger assembly that allowed the operator to position the suppository reproducibly. The remainder of the chamber was filled with glass beads, and its upper section was connected. The chamber then was lowered into the water bath along with the glass coil and fixed into an upright position. The pump was started immediately in the reverse direction, allowing the water to fill the test chamber from the bottom up. As soon as the entire system was filled, the flow was reversed so that the water flowed from top to bottom in the chamber.

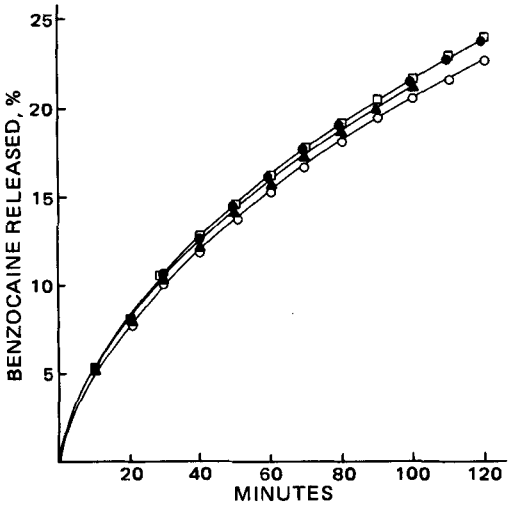


Figure 5—*Replicate 37° dissolution experiments for the release of benzocaine from Base B using the modified dissolution cell. Key: ○ and □, Day 1; and ▲ and ●, Day 2.*

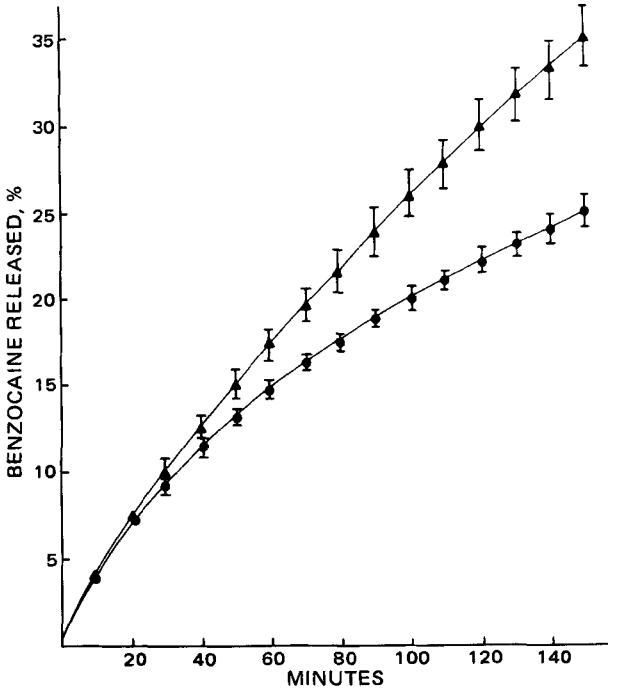


Figure 6—*Comparative release of benzocaine from Base A (▲) and B (●) suppositories at 37°.*

The flow rate was controlled at 30 ± 0.5 ml/min. Drug release was monitored with a UV spectrophotometer (282 nm) equipped with a recorder. The cumulative amount released with time was calculated from the recorder tracings by converting absorbance to concentration and multiplying by the volume of the dissolution medium. Before each run, the UV spectrophotometer was checked with two solutions of known benzocaine concentrations to assure that absorbance was linear with concentration.

RESULTS AND DISCUSSION

The concept of a flow system employing a bed of beads was shown to be useful for studying dissolution rates of solids. A system for testing tablets and granules in which the sample is placed within the bed was described previously (49). Reproducible dissolution rates for drug powders were obtained (50) by mixing the powder with a diluent and placing the mixture between two layers of glass beads in a flowcell. Rippie and Huq (51) used a flowcell containing several layers of glass beads in which the bead size was reduced successively. Drug powder, introduced at the top, then migrated through the bed as dissolution occurred. These recent reports demonstrate the distinctness of this type of system opposed to the usual vessel and flow systems used for dissolution testing.

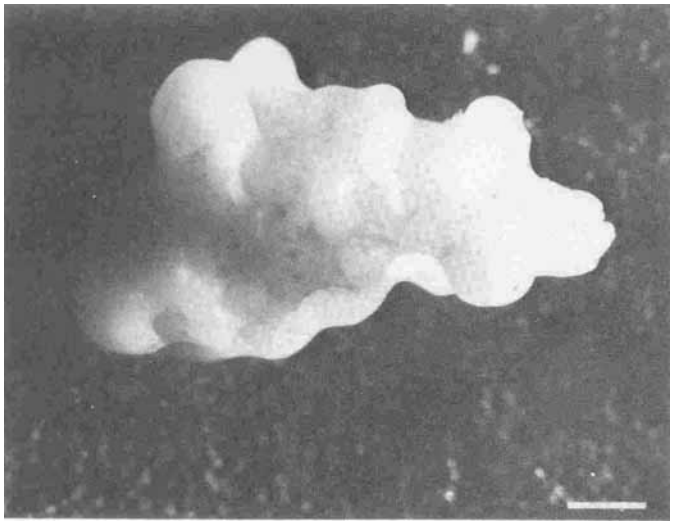
Suppository drug release is influenced by factors such as: drug–vehicle interactions; vehicle composition; crystalline form of the vehicle (polymorph); solubility, partition coefficient, and drug permeability in the vehicle; particle size of the drug (dispersed system); and drug concentration in the vehicle. Experimental parameters such as the temperature of the dissolution solvent and flow rate should influence the observed release patterns.

This report presents a preliminary evaluation of the continuous flow bead-bed concept using two types of suppository bases with benzocaine as the model compound. Benzocaine was selected because benzocaine suppositories are marketed and because it has a UV-absorbing chromophore that allows it to be easily assayed.

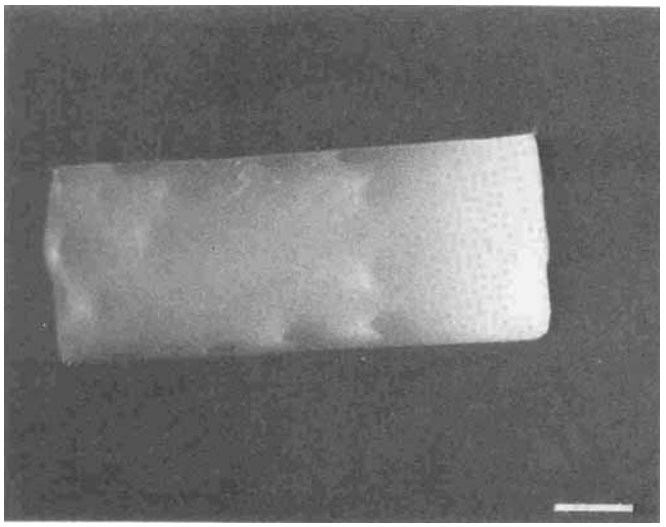
The two suppository bases (A and B) were mixtures of glycerides of fatty acids. They possess physicochemical properties representative of the many vehicles purchased from commercial suppliers (10) and have been studied with several different prostaglandins (3–5).

Figures 3 and 4 show the release profiles of benzocaine in duplicate from Bases A and B, respectively, on each of 2 different days, using the original dissolution chamber. Figure 5 gives the release profiles from the new chamber. These data indicate that there was no major difference between the two chambers (*i.e.*, within 10%).

The replicate runs were performed within 3 days to avoid any potential effects of a polymorphic change of the base on the drug release rate. The



BASE A



BASE B

Figure 7—*Photographs of suppositories after 2 hr (37°) in the bead-bed dissolution apparatus. Scale marker = 0.31 cm.*

thermal behavior of Base B (with benzocaine) was verified by differential scanning calorimetry; these scans were invariant with time[7]. The dissolution results indicate very good reproducibility of the data. Comparisons of the 15 time points on the curves from four runs gave an average relative standard deviation of 6.13% for Base A and 3.36% for Base B. Therefore, 95% of the time the complete release profile will be reproduced with a 13% variation for Base A and a 7% variation for Base B.

Release from the bases is compared in Fig. 6. The bars represent the standard deviation of the points. The percentages of benzocaine released during the first 30 min were similar, but the lower melting Base A exhibited a much faster release at later times. Figure 7 shows the physical form of the two bases after 2 hr in the bead-bed at 37°. They were allowed to solidify at room temperature before removal.

The low melting Base A softened and stayed within a confined region of the bead-bed. The higher melting Base B maintained its form, showing slight indentations from the beads. Suppositories of these types do not show complete melting at 37° but rather a softening of the suppository matrix. At temperatures well above their melting points, total liquefaction takes place. The degree to which the beads actually penetrate the suppository depends on the thermal behavior of a given suppository.

The similar early release of benzocaine from the two bases most likely results from the fact that they initially are both rigid. It requires time

[7] After ~1 month of storage at 25°, the differential scanning calorimetric curves began to change with a concomitant decrease in the release profiles. Alteration in the thermal behavior of semisynthetic bases was recently reported (52).

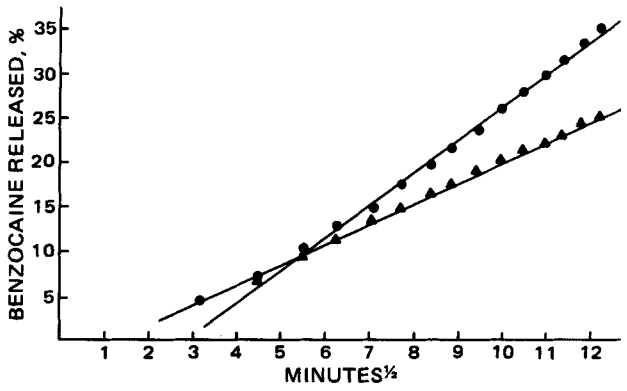


Figure 8—*Release of benzocaine, plotted as the square root of time, from Base A (●) and B (▲) suppositories at 37°.*

before the suppositories soften sufficiently to detect significant differences in release. Visual examination of Base A after 5 and 20 min supported this view. After this time, bead penetration restricts the area for drug release. Therefore, *in vitro* release from Base A is actually higher than observed. Plots of the percent released *versus* the square root of time were linear with correlation coefficients of 0.990 and 0.996, respectively (Fig. 8). This type of release kinetics follows the matrix-controlled release theory previously developed (53). It is particularly applicable to Base B, which maintained its shape (constant surface area) during the test period.

To investigate bead penetration further, the influence of temperature on the release of benzocaine from Base B was studied in more detail. Release curves as a function of temperature are presented in Fig. 9. The release profiles did not increase as a direct function of temperature. For example, release increased from 33 to 37°, with 39 and 40° data being comparable to the 33° run.

The fastest release profile was achieved at 45° where complete melting of the base occurred. At first, this overall temperature dependence seemed surprising. However, close examination of the physical state of the suppository in the beads provided the reason for this behavior. While the suppository was in the dissolution apparatus, it softened and eventually melted. As it softened, the beads became impressed within the suppository (Fig. 10). At 39 and 40°, the beads penetrated the surface of the suppository, thereby reducing the surface area available for drug release. Once the base melted completely (*e.g.*, 45°), release increased again due to the spreading of the oil in the bead-bed. At this temperature, oil globules were found floating at the top of the beads. Although release was temperature dependent, it was reproducible at the temperatures studied. This finding supports the continued evaluation and development of this apparatus, which is particularly well suited for suppositories that reasonably maintain their shape during the dissolution process.

In view of the influence of the beads on release at temperatures that

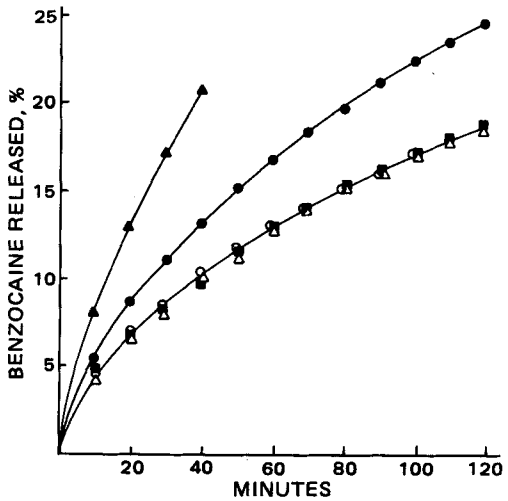


Figure 9—*Dependence of benzocaine release from Base B suppositories as a function of temperature. Key: ▲, 45°; ■, 40°; ○, 39°; ●, 37°; and △, 33°.*

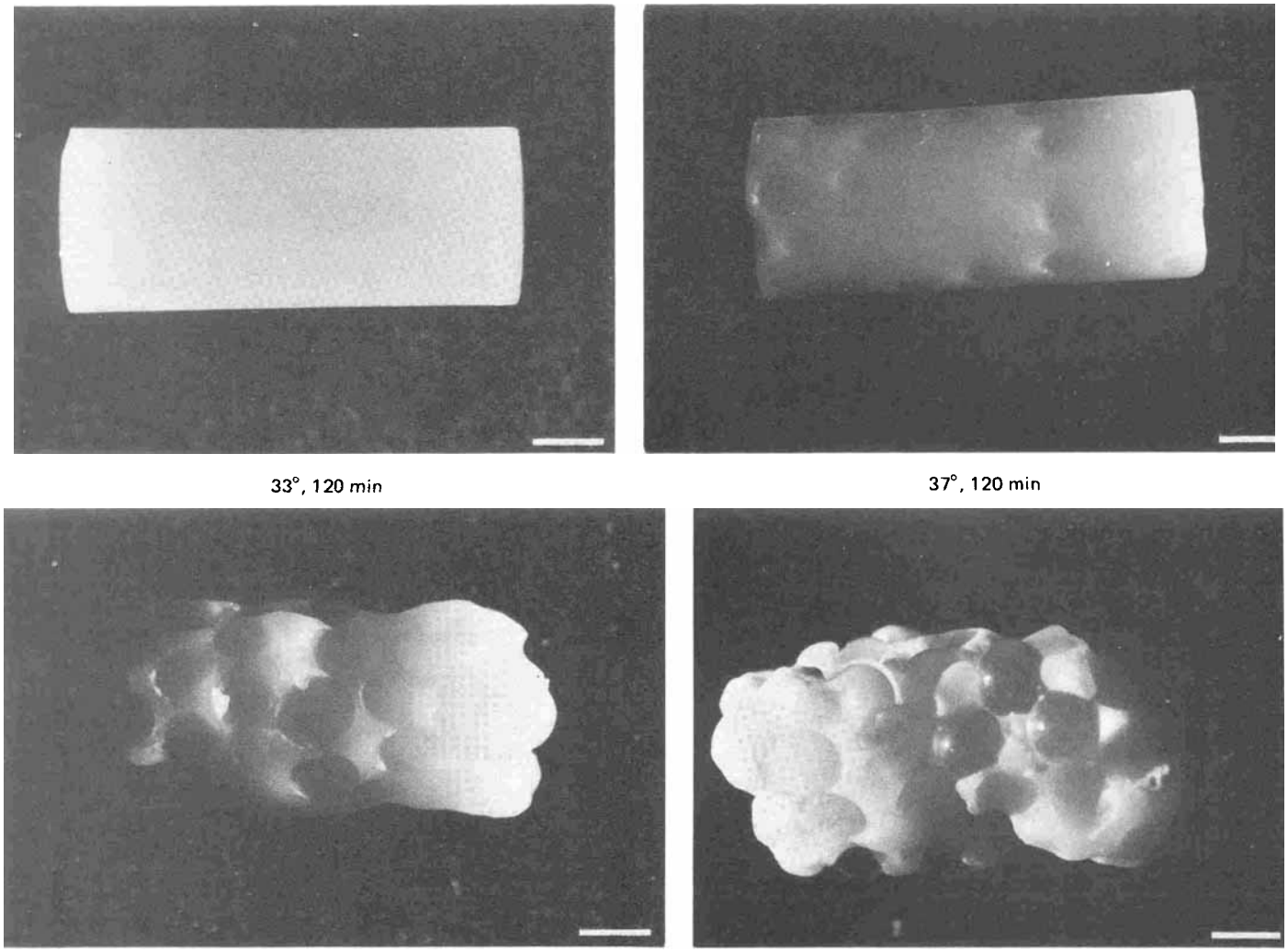


Figure 10—*Photographs of Base B suppositories after ~2 hr (37°) in the bead-bed dissolution apparatus. Scale marker = 0.31 cm.*

cause deformation of the base, further refinements of the bead-bed apparatus are anticipated. Consideration will be given to maintaining the beads in a stationary position, thereby eliminating the dynamics of bead penetration. This can be accomplished by using a preformed bead cavity as a mold or a porous gel instead of the beads.

REFERENCES

(1) "Sprowl's American Pharmacy," L. W. Dittert, Ed., Lippincott, Philadelphia, Pa., 1974.

(2) T. O. Oesterling, W. Morozowich, and T. J. Roseman, *J. Pharm. Sci.*, **61,** 1861 (1972).

(3) M. Mandelin, *Prostaglandins*, **16,** 143 (1978).

(4) M. Bygdeman, J. N. Martin, A. Leader, V. Lundstrom, M. Ramadan, P. Eneroth, and K. Green, *Obstet. Gynecol.*, **48,** 221 (1976).

(5) N. H. Lauersen and K. H. Wilson, *Prostaglandins*, **10,** 1037 (1975).

(6) F. A. Kimball, G. L. Bundy, A. Robert, and J. R. Weeks, *ibid.*, **17,** 657 (1979).

(7) V. Lundstrom, M. Bygdeman, S. Fotiou, K. Green, and K. Kinoshita, *Contraception*, **16,** 167 (1977).

(8) "Obstetric and Gynaecological Uses of Prostaglandins," S. M. M. Karim, Ed., Asian Federation of Obstetrics and Gynaecology, 1st Inter-Congress, Singapore, Apr. 27–30, 1976.

(9) T. J. Roseman, G. D. Gutknecht, R. G. Stehle, and E. M. Southern, *Am. J. Obstet. Gynecol.*, **129,** 225 (1977).

(10) "The Theory and Practice of Industrial Pharmacy," L. Lachman, H. A. Lieberman, and J. L. Kanig, Eds., Lea & Febiger, Philadelphia, Pa., 1970.

(11) C. W. Whitworth and J. P. LaRocca, *J. Am. Pharm. Assoc., Sci. Ed.*, **48,** 353 (1959).

(12) A. J. M. Schoonen, F. Moolenaar, C. Haverschmidt, and T. Huizinga, *Pharm. Weekbl.*, **111,** 585 (1976).

(13) H. M. Gross and C. H. Becker, *J. Am. Pharm. Assoc., Sci. Ed.*, **42,** 96 (1953).

(14) S. N. Pagay, R. I. Poust, and J. L. Colaizzi, *J. Pharm. Sci.*, **63,** 44 (1974).

(15) P. Vaghy and P. Vaghy, *Gyogyszereszet*, **22,** 27 (1978).

(16) C. J. deBlaey and J. J. Rutten-Kingma, *Pharm. Acta Helv.*, **52,** 11 (1977).

(17) I. W. Kellaway and C. Marriott, *J. Pharm. Sci.*, **64,** 1162 (1975).

(18) V. E. Krogerus and M. Tolvi, *Acta Pharm. Suec.*, **2,** 327 (1965).

(19) E. L. Parrott, *J. Pharm. Sci.*, **64,** 878 (1975).

(20) L. Roller, *Aust. J. Hosp. Pharm.*, **7,** 97 (1977).

(21) S. E. Leucuta, L. Popa, M. Ariesan, L. Popa, R. D. Pop, M. Kory, and S. Toader, *Pharm. Acta Helv.*, **52,** 261 (1977).

(22) A. Puech, Y. Lasserre, and M. Jacob, *Trav. Soc. Pharm. Montpellier*, **37,** 165 (1977).

(23) L. Bardet and J. Cemeli, *ibid.*, **16,** 200 (1956).

(24) V. P. Bhavnagri and P. Speiser, *Pharm. Acta Helv.*, **51,** 10 (1976).

(25) M. A. Ghafoor and C. L. Huyck, *Am. J. Pharm.*, **134,** 63 (1962).

(26) A. A. Kassem, E. N. El-Din, A. A. El-Bary, and H. M. Fadel, *Pharmazie*, **30,** 472 (1975).

(27) L. Krowczynski, *Acta Pol. Pharm.*, **19,** 127 (1962).

(28) Z. Kubiak, *Gyogyszereszet*, **21,** 322 (1977).

(29) V. H. Muhlemann and R. H. Neuenschwander, *Pharm. Acta*

Helv., **31**, 305 (1956).
(30) M. Kapas, E. Regdon, and G. Regdon, *Acta Pharm. Technol.*, **25**, 109 (1979).
(31) G. Regdon, A. Magyarlaki, G. Kedvessy, E. Minker, and E. Regdon, *Pharmazie*, **33**, 67 (1978).
(32) R. Voigt and G. Falk, *ibid.*, **23**, 709 (1968).
(33) G. B. Carp, D. Brossard, C. Chemtob, and J. C. Chaumeil, *Sci. Tech. Pharm.*, **7**, 159 (1978).
(34) W. H. Thomas and R. McCormack, *J. Pharm. Pharmacol.*, **23**, 490 (1971).
(35) T. Stozek, *Pol. J. Pharmacol. Pharm.*, **27**, 227 (1975).
(36) L. Turakka and V. E. Krogerus, *Farm. Aikak.*, **83**, 59 (1974).
(37) *Ibid.*, **83**, 105 (1974).
(38) J. M. Plaxco, Jr., C. B. Free, Jr., and C. R. Rowland, *J. Pharm. Sci.*, **56**, 809 (1967).
(39) J. W. Ayres, D. Lorskulsint, A. Lock, L. Kuhl, and P. A. Laskar, *ibid.*, **65**, 832 (1976).
(40) C. G. Hartman, *Ann. N.Y. Acad. Sci.*, **83**, 318 (1959).
(41) H. P. M. Kerckhoffs and T. Huizinga, *Pharm. Weekbl.*, **102**, 1187, 1255 (1967).
(42) C. F. Peterson and A. J. Guida, *J. Am. Pharm. Assoc., Sci. Ed.*, **42**, 537 (1953).
(43) H. Piasecka and Z. Zakrzewski, *Pol. J. Pharmacol. Pharm.*, **28**, 199 (1976).
(44) W. A. Ritschel and M. Banarer, *Arzneim.-Forsch.*, **23**, 1031 (1973).
(45) S. Tsuchiya, M. Hiura, and H. Matsumaru, *Chem. Pharm. Bull.*, **25**, 667 (1977).
(46) W. A. Ritschel and J. Rotmensch, *Pharm. Int.*, **3**, 4 (1974).
(47) M. R. Baichwal and T. V. Lohit, *J. Pharm. Pharmacol.*, **22**, 427 (1970).
(48) H. W. Puffer and W. J. Crowell, *J. Pharm. Sci.*, **62**, 242 (1973).
(49) U. Bogs and A. Darr, *Pharm. Praxis*, **31**, 169 (1976).
(50) F. Langenbucher and H. Rettig, *Drug Dev. Ind. Pharm.*, **3**, 241 (1977).
(51) E. G. Rippie and A. Huq, *J. Pharm. Sci.*, **68**, 938 (1979).
(52) L. J. Coben and N. G. Lordi, *ibid.*, **69**, 955 (1980).
(53) T. Higuchi, *ibid.*, **52**, 1145 (1963).

Effect of Caffeine on Ergotamine Absorption from Rat Small Intestine

JOHN R. ANDERSON, GERTRUDE DREHSEN, and IAN H. PITMAN ˣ

Received July 24, 1980, from the *Victorian College of Pharmacy Ltd., Parkville, Victoria, Australia 3052.* Accepted for publication December 2, 1980.

Abstract □ The effect of caffeine on the absorption of ergotamine from the rat small intestine was studied. The results of a series of experiments showed that caffeine significantly enhanced absorption of ergotamine from solutions of pH 5.0 when both substances were in solution and when an intact blood supply was either absent (*in vitro* everted sac experiments) or present (*in situ* experiments). Caffeine did not appear to influence the absorption rate of ergotamine in *in situ* experiments when the solution pH was 3.0. Isosorbide dinitrate, a vasodilator, enhanced ergotamine absorption when both substances were administered simultaneously into intestinal loops *in situ*. Isosorbide dinitrate probably exerts its effect by increasing blood flow to the intestine. The results are consistent with an hypothesis that the rate-determining step in ergotamine absorption is the transport of the drug from a lipid phase (GI membrane) into an aqueous phase (blood). Caffeine is thought to exert its rate-accelerating effect by increasing the water solubility of ergotamine neutral molecules.

Keyphrases □ Caffeine—effect on ergotamine absorption from rat small intestine, *in vitro* and *in vivo* studies □ Ergotamine—effect of caffeine on absorption from rat small intestine, *in vitro* and *in vivo* studies □ Absorption—ergotamine, effect of caffeine on absorption from rat small intestine, *in vitro* and *in vivo* studies □ Analgesics—ergotamine, effect of caffeine on absorption from rat small intestine, *in vitro* and *in vivo* studies

Ergotamine tartrate is widely used in the treatment of migraine (1, 2). When administered parenterally, it is effective in relieving migraine pain (3, 4), but oral administration often affords no relief (5, 6). Regardless of the route, ergotamine must be administered early in an attack to be effective (7). The frequent failure of oral ergotamine therapy appears to be related to its relatively low water solubility (1:500) and the fact that its absorption from the GI tract into the systemic circulation often is slow or impaired during a migraine attack (4–9).

Simultaneous oral administration of caffeine and ergotamine results in more effective therapy than ergotamine administered alone (10–12). Recent pharmacokinetic studies in humans indicated faster and more complete absorption of ergotamine after oral administration when it is combined with caffeine (13). Zoglio *et al.* (14) conducted *in vitro* experiments to elucidate the mechanism by which caffeine enhances ergotamine absorption; caffeine increased the solubility and dissolution rates of ergotamine in 0.1 *M* HCl and in 0.1 *M* phosphate buffer (pH 6.65) and was claimed to accelerate the partitioning of ergotamine from an aqueous (pH 6.65) to an organic (chloroform) phase. These observations led to the conclusion that the solubilizing effect of caffeine on ergotamine in water is the major factor in its enhancement of oral ergotamine migraine therapy.

The present study was undertaken to determine the effect, if any, that caffeine has on ergotamine absorption across the rat small intestine when both substances are dissolved.

EXPERIMENTAL

Ergotamine Assay—Ergotamine concentrations were calculated from the results of reversed-phase high-performance liquid chromatographic (HPLC) assays (15). The mobile phase was acetonitrile–1% sodium acetate/acetic acid buffer (55:45) at pH 6.5. The stationary phase was a 10/25 ODS column, and ergotamine was detected by fluorescence spectrometry (λ_{ex} = 325 nm; λ_{em} = 407 nm).

***In Vitro* Studies (Everted Sac Technique)**—Adult male Hooded Wistar rats, 240–280 g, were fasted in cages designed to prevent coprophagy for at least 12 hr before sacrifice. Water was allowed *ad libitum*. The rats were killed by a blow on the head. The small intestine was removed immediately, rinsed with Krebs–Henselite solution, sleeved onto a glass rod, and everted carefully. The first 15 cm, beginning with the pylorus, was discarded, and the next 8-cm segment was used in the experiments.

The segment was attached to a cannula at the proximal end, trimmed to 7 cm, and ligated at the distal end so that 6 cm of the everted intestine

0022-3549/81/0600-0651$01.00/0