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Dosage Form Design for Improvement of Bioavailability of Levodopa VI: Formulation of Effervescent Enteric-Coated Tablets

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Received March 4, 1983, from the *Product Development Laboratories, Sankyo Company, Ltd., 1-chome, 2-58, Hiromachi, Shinagawa, Tokyo, Japan.* Accepted for publication July 1, 1983.

Abstract □ A new dosage form of levodopa, which has the characteristics of loading high concentrations of levodopa at the upper part of the intestine, has been developed to improve its bioavailability. It is shown that an effervescent tablet formulation, coated with hydroxypropyl methylcellulose phthalate (carboxybenzoyl radical content: 20–24%) as the enteric material, is suitable for the purpose of dissolution. This was confirmed from animal experiments, which showed that tablets of this composition disintegrate instantly on reaching the upper part of the intestine. This tablet was considered appropriate for the bioavailability tests described in this paper.

Keyphrases □ Levodopa—effervescent enteric-coated tablet, intestinal absorption, dissolution □ Effervescent enteric-coated tablet—levodopa, intestinal absorption, lag time of dissolution and absorption, effect of size and shape □ Film material—hydroxypropyl methylcellulose phthalate, effervescent enteric-coated tablet, levodopa

It has been shown in previous work in this series (1, 2) that the bioavailability of levodopa could be improved by loading high concentrations of the drug at the upper part of the intestine, the optimum site of absorption, and then inducing temporary saturation of levodopa decarboxylase. The present work describes the preparation of an oral solid dosage form of levodopa with improved bioavailability. The *in vitro* dissolution and *in vivo* disintegration behavior also are reported.

EXPERIMENTAL SECTION

Preparation of Effervescent Enteric-Coated Tablets of Levodopa [I]—A granular mixture of levodopa USP and carboxymethylcellulose[1] was prepared by the wet granulation method using an aqueous solution of hydroxypropyl cellulose[2] as a binder. All other diluents, effervescent agents, lubricants, and/or coloring agents were mixed with the dried granulation, and the mixture was compressed into tablets. These tablets were coated successively with hydroxypropyl methylcellulose USP (II) (5% w/w) and then with hydroxypropyl methylcellulose phthalate (III)[3] (10% w/w) in methylene dichloride–ethanol (1:1, w/w) solution[4] to obtain enteric-coated tablets. All the coating agents that were used here are accepted for drug use by the FDA.

Dissolution Tests—A modified disintegration test apparatus was used in the dissolution tests of levodopa and/or dyes (3). The frequency rate of the basket-rack assembly was set between 5 and 20 cpm. Ten-mesh stainless steel cloth was fitted over the top of the basket-rack assembly to prevent the tablet from floating out of the tube of the assembly during dissolution.

Dissolution media (900 mL) at various pH values, prepared by mixing test solutions 1 and 2 (JP IX), were heated to 37 ± 0.5°C and placed in the dissolution apparatus. One tablet was placed in the basket. The solution was drawn from the flask through the flow cell (length 5 mm) by a pump[5] and returned to the flask at a flow rate of ~25 mL/min. The differences in absorbance between λ_1 (258.5 nm) and λ_2 (281 nm) for levodopa, λ_1 (620 nm) and λ_2 (662 nm) for methylene blue, and λ_1 (600 nm) and λ_2 (522 nm) for erythromycin were measured with a dual-wavelength spectrophotometer[6] and

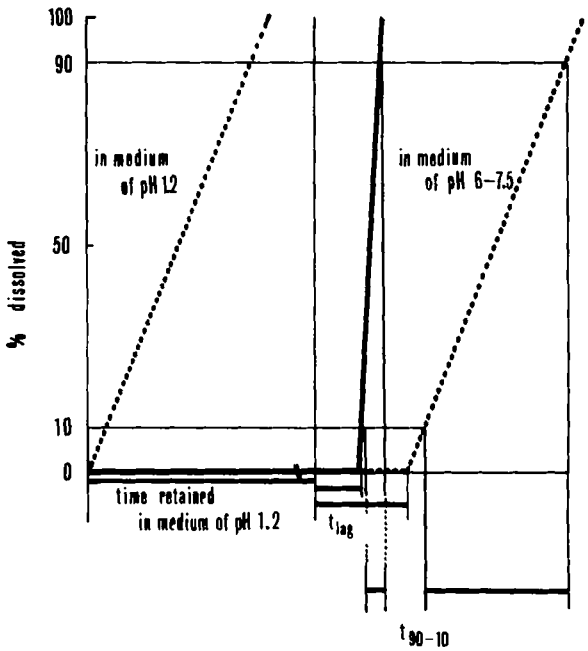

Figure 1—*Dissolution patterns of levodopa. The solid line shows the desired pattern; the dotted lines show conventional dosage forms.*

[1] Marketed as NS300, Gotoku Yakuhin, Tokyo, Japan; acid form of sodium carboxymethylcellulose USP.
[2] Nippon Soda Co., Ltd., Oiso-machi, Kanagawa, Japan; F.C.C. III P280.
[3] Shinetsu Chemicals Co., Ltd., Tokyo, Japan; Biddle Sawyer Corporation, 2, Penn-Plaza, New York, N.Y. 10121, U.S.A.; Master File No. DMF-2151.
[4] A substitute solution is acetone–ethanol solution (1:1, w/w) or acetone–isopropyl alcohol (1:1, w/w).
[5] Type MBP-100, Iwaki Co., Ltd., Japan.
[6] Hitachi-156 Spectrophotometer; Hitachi Co., Ltd., Tokyo, Japan.

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Table I—Formulations of Effervescent Enteric Tablets of Levodopa for the Tests of Dissolution Parameters

Compound	Amount per Tablet, mg				
	1	2	3	4	5
Levodopa	200	200	200	200	200
Tartaric acid	0	6.25	12.5	25	50
Sodium bicarbonate	0	7	14	28	56
Lactose	106	92.75	79.5	53	0
Carboxymethylcellulose, Magnesium aluminum silicate, Sodium bisulfate, Hydroxypropyl cellulose, Magnesium stearate	79	79	79	79	79
Hydroxypropyl methylcellulose	8	8	8	8	8
Hydroxypropyl methylcellulose phthalate-50	30	30	30	30	30
Total weight	423	423	423	423	423

recorded continuously. The following parameters were obtained from the dissolution curves in several runnings: t_{lag} (lag time of dissolution), t_{10} (time required to dissolve 10% of labeled drug), t_{90} (time required to dissolve 90% of labeled drug), and $t_{90} - t_{10}$.

Measurement of the Dissolution Time of the Enteric Coating Material—Fifty-milligram portions of 20-32-mesh fractions of each coating material were suspended in media of various pH values, as described in the dissolution test, and agitated with a magnetic stirrer (9 mm in diameter and 30 mm in length) at ~500 rpm at 37 ± 0.5°C. The dissolution time of the suspended particles was measured visually.

Lag Time Determination of Levodopa Absorption—Two samples were used for this study. Ten healthy male beagle dogs, 10.0–14.0 kg, were fasted for ~16 h and divided into two groups of five dogs each. Crossover experiments were carried out at 1-week intervals. The dogs were dosed orally, and levodopa absorption was measured by observing the time at which the dogs began vomiting, which indicated a significant release of the drug with subsequent absorption.

Measurement of the Disintegration Site of Tablets *In Vivo*—*Experiment No. 1*—Two male beagle dogs, 10.7 and 11.3 kg, were each orally dosed with two samples. Immediately after the dogs began vomiting, they were anesthetized with pentobarbital sodium[7] (0.6 mL/kg), placed on their backs, and killed by exsanguination. The abdomens were immediately opened and the entire GI tracts were excised and opened in order to observe the disintegration sites. Such observation was made possible by the dye contained in the tablets.

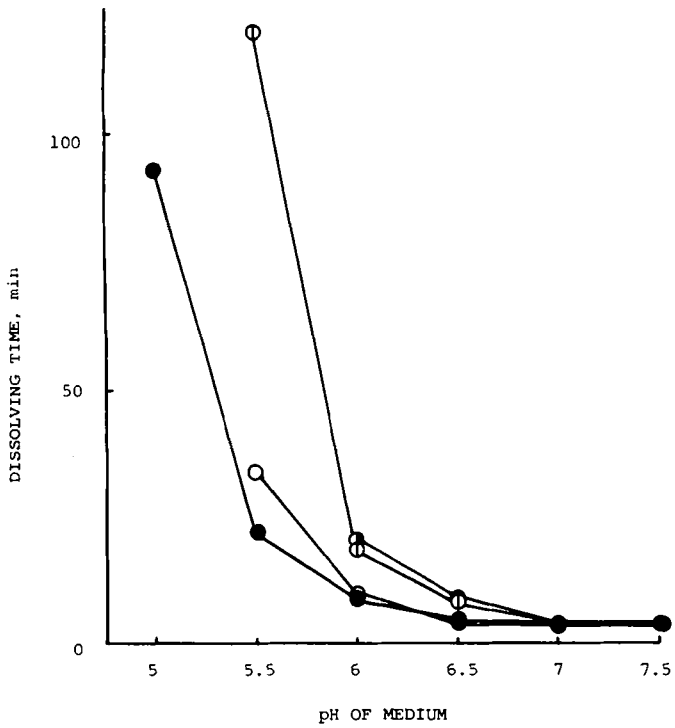

Figure 2—*pH profile of dissolution time of various enteric film materials. Key: (●) III-45; (○) III-50; (◐) III-55; (◑) IV.*

[7] Dainippon Pharmaceutical Co., Ltd.

Experiment No. 2—One male beagle dog, 13.9 kg, was orally dosed with two tablets from each sample. Fifty minutes after the dog began vomiting, it was anesthetized with pentobarbital sodium 6 and examined, as in experiment no. 1.

RESULTS AND DISCUSSION

From the results of the *in vitro* and *in situ* experiments of levodopa absorption characteristics, it was found that levodopa absorption from the solid dosage form was enhanced by:

1. Selecting the correct enteric dosage form to inhibit the levodopa decomposition in the stomach and ensure a high concentration of levodopa at the absorption site.
2. Selecting the appropriate enteric film materials to obtain rapid disintegration and dissolution characteristics in the upper part of the intestine.
3. Investigating dissolution characteristics so as to obtain a high concentration of levodopa at the onset of dissolution and to inhibit the conversion to dopamine (4) as much as possible by decarboxylase distributed in the intestine, during the absorption process.
4. Investigating product form and size to minimize variations in lag time of absorption and to shorten the lag time of absorption due to variability of individual gastric transit times.

The *in vitro* dissolution curve showing the aforementioned characteristics can be seen in Fig. 1. One of the main purposes of this report was to obtain a preparation having the dissolution characteristics shown in Fig. 1.

Selection of Enteric Film Material—The hydroxypropyl methylcellulose phthalate (III) used had different carboxybenzoyl radical contents (45[8], 50[9], 55[10]). Cellulose acetate phthalate (IV) was selected as a reference because

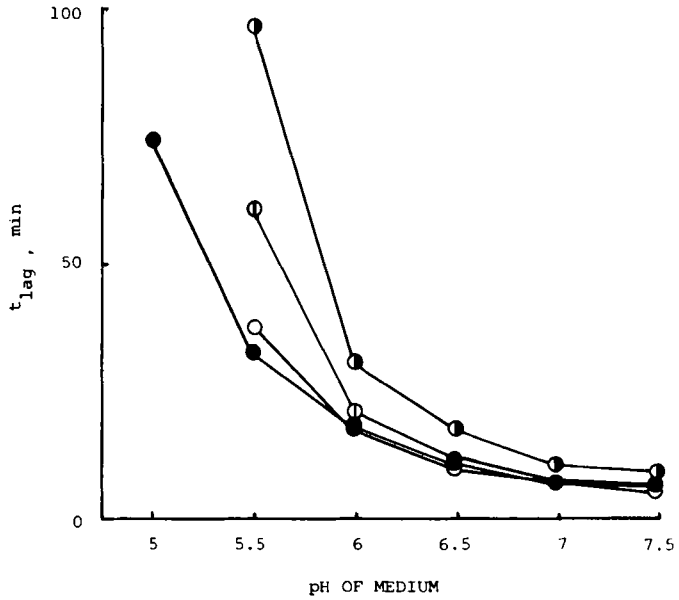

Figure 3—*pH profile of the dissolution parameter, t_{lag}, of effervescent enteric tablets of levodopa coated with various enteric film materials. Key: (●) III-45; (○) III-50; (◐) III-55; (◑) IV.*

[8] Carboxybenzoyl radical content: ~20%.
[9] Carboxybenzoyl radical content: 20–24%.
[10] Carboxybenzoyl radical content: 27–35%.

Table II—Formulations and Dissolution Parameters of Two Samples Employed for the Determination of Disintegration Sites *In Vivo*

Compound	Amount per Tablet, mg A	B
Levodopa	180	180
Methylene blue	20	0
Erythromycin	0	20
Tartaric acid	25	25
Sodium bicarbonate	28	28
Carboxymethylcellulose, Microcrystalline cellulose, Magnesium aluminum silicate, Sodium bisulfate, Hydroxypropyl cellulose, Magnesium stearate	79	79
Hydroxypropyl methylcellulose	8	8
Hydroxypropyl methylcellulose phthalate-50	30	0
Cellulose acetate phthalate	0	30
Total weight	370	370

	Dissolution Parameter, min	
Levodopa		
pH 7.5		
t_{lag}	5.3	7.0
$t_{90\text{-}10}$	1.1	1.5
pH 6.0		
t_{lag}	18.3	36.2
$t_{90\text{-}10}$	2.0	2.1
Dye		
pH 7.5		
t_{lag}	5.6	8.6
$t_{90\text{-}10}$	1.4	1.1
pH 6.0		
t_{lag}	22.7	35.1
$t_{90\text{-}10}$	1.5	1.7

it is used extensively in the marketplace. The relationship between the dissolution time of the enteric film material and the pH of the medium is shown in Fig. 2. It can be seen from Fig. 2 that there is no difference in dissolution time between materials at pH 7.0 and at pH 7.5. However, at more acidic pH levels, the dissolution time of the material was longer in the order of III-45, III-50, III-55, and IV. The relationship between dissolution lag time (t_{lag}) of tablets coated with various enteric materials and medium pH was also determined (Fig. 3), from which the same pattern as the film materials themselves can be seen.

Intestinal pH is known to be between ~6 and 7.5. It has been reported, however, that the pH of the duodenum was ~pH 7 and varied according to the pH of the stomach (5). On the other hand, it has been reported that disintegration and dissolution behavior at more acidic medium levels than the disintegration test solution (pH 7.5) should be used in evaluating bioavail-

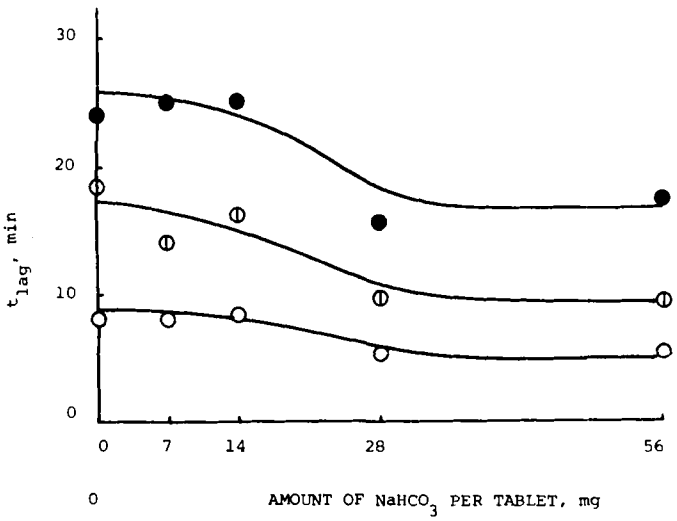

Figure 4—*Relationship between the dissolution parameter, t_{lag}, of effervescent enteric tablets of levodopa and the amount of sodium bicarbonate formulated in the tablet. Key: (●) pH 6.0, 20 strokes/min; (○) pH 7.5, 5 strokes/min; (⊖) pH 7.5, 20 strokes/min.*

Table III—*In Vivo* Disintegration of Samples A and B

	Dog 1	Dog 2	Dog 3
Body weight, kg	10.7	11.3	13.9
Length of intestine, cm	170	180	170
Length of duodenum, cm	20	25	25
Time of emesis, min postdose	120	110	130
Anesthetizing time, min postdose	120	110	180
Results from Sample A			
Dose, number of tablets	1	1	2
Appearance of tablets	d.c.[a]	d.c.	d.c.
Distance from pylorus, cm	20	23	20
Results from Sample B			
Dose, number of tablets	1	1	2
Appearance of tablets	n.d.[b]	n.d.	d.c.
Distance from pylorus, cm	50	60	40, 130

[a] Disintegrated completely. [b] Not disintegrated.

ability of enteric-coated tablets (6). Moreover, considering the prevention of disintegration in the stomach, it might be more appropriate to evaluate film dissolving time or lag time of dissolution of enteric-coated tablets at pH 6–6.5. Therefore, III-50 was selected as the most appropriate material from among the four kinds of films used.

Preparation of I—Effervescent tablets were investigated to obtain characteristics of fast disintegration and instantaneous dissolution of levodopa preparations. The compressibility of levodopa was such as to require additives. For this reason, it was difficult to obtain the characteristics mentioned above in other dosage forms. A sodium bicarbonate–tartaric acid system was selected as the effervescent component, and the five kinds of tablets shown in Table I were prepared.

For these tablets, the relationship between the amount of effervescent component and lag time of dissolution (t_{lag}) and between the amount of effervescent component and rate of dissolution ($t_{90\text{-}10}$) were studied. The results are shown in Figs. 4 and 5. In the determination of $t_{90\text{-}10}$, the frequency of the basket-rack assembly was set to 5 cpm to clarify the differences among the preparations. It can be seen from Fig. 4 that as the amount of effervescent component is increased, the dissolution lag time will decrease. However, the dissolution lag time was found to become constant, and as seen in Fig. 5, the dissolution rate became smaller with the addition of >28 mg/tablet of sodium bicarbonate. These results confirmed, therefore, that formulation 4 in Table I was the more appropriate.

It is difficult to say why the lag times are different among the preparations,

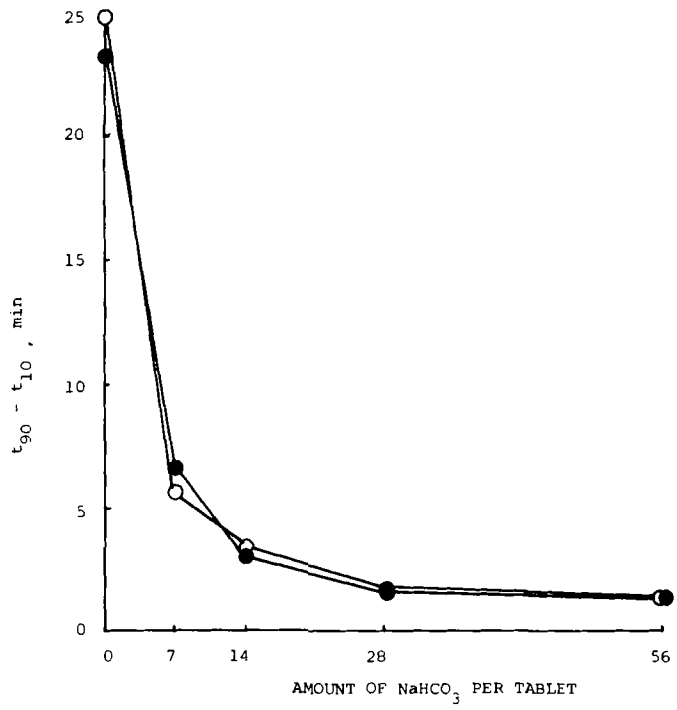

Figure 5—*Relationship between the dissolution parameter $t_{90\text{-}10}$ of effervescent enteric tablets of levodopa and the amount of sodium bicarbonate formulated in the tablet. The number of strokes was fixed at 5/min, and the pH was 7.5. Key: (●) uncoated tablet; (○) enteric tablet.*

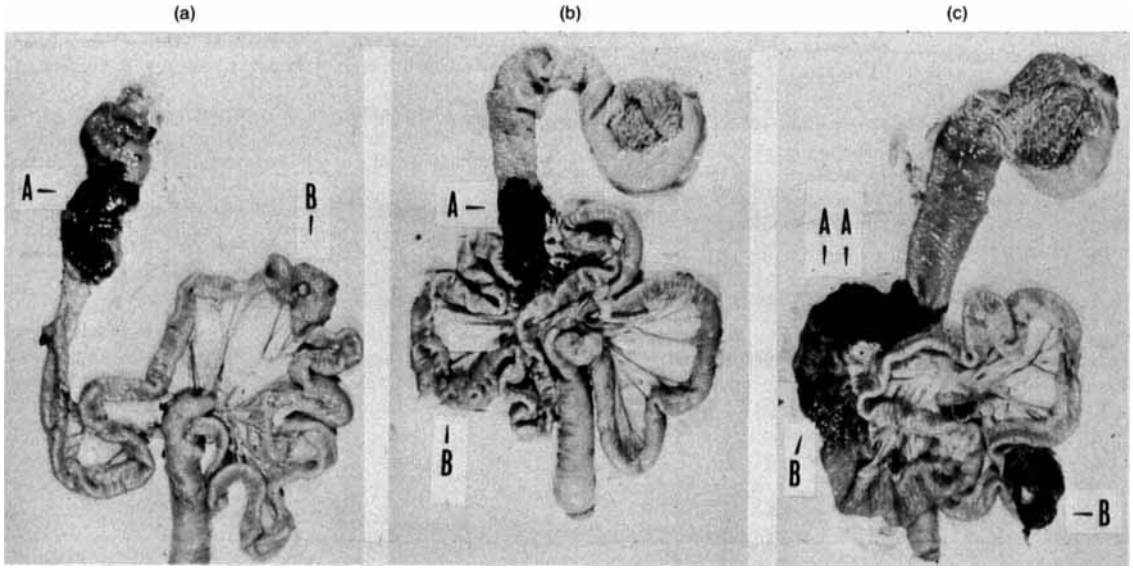

Figure 6—*Photographs of the* in vivo *disintegration site of samples A and B. Key: (a) dog 1; (b) dog 2; (c) dog 3. See Table III for experimental details.*

even with the same enteric film. One reason may be the variability in the rate of evolution of carbon dioxide due to slight differences in the rate of penetration of water into the tablet.

Disintegration Site of I—The disintegration site of I was investigated in beagle dogs to confirm *in vivo* behavior. Two samples, A and B, were used in the experiments; compositions and dissolution parameters are shown in Table II. Dyes were added to facilitate visual investigation of the site of and the rate of disintegration. Results are summarized in Table III and intestinal dissections are shown in Fig. 6.

The measured lengths of the duodenum and the small intestine are approximate. The lengths of the duodena measured were the same as that described in the literature (7) (*i.e.*, ~25 cm). As shown in Table III and Fig. 6, sample A disintegrated in the duodenum of each dog and the length of the dyeing site was short. These results suggest that disintegration of I started as soon as it reached the intestine and that the rate of disintegration or dissolution was instantaneous. In contrast, disintegration of sample B was very slow, and the sites of the disintegrations varied between the two tablets. In comparison with the dissolution parameters shown in Table III, it seems reasonable to assume that the dissolution lag time is correlated with the site of disintegration and that the rate of dissolution ($t_{90\text{-}10}$) is correlated with the length of the dyed site. Tablet I was designed for fast disintegration and high concentration in the upper intestine. From the above results, it was concluded that sample A was most suited to the purpose.

Effects of Tablet Shape and Size on the Lag Time of I—A 250-mg volume of levodopa was formulated into oval and round effervescent enteric-coated tablets. Table IV shows the characteristics of the two tablets. The tablets were compared with respect to lag time of levodopa absorption in dogs. The results (Fig. 7) indicate that the round tablet gives a shorter lag time and smaller intersubject variation in comparison with the oval one.

Three types of I, each containing 200 mg of levodopa but of different sizes, were prepared. Table IV shows the characteristics of the three sizes of tablets. Lag times of levodopa absorption were determined following oral administration of four kinds of I: the round tablet and the three types of tablets (samples 1–3) in Table IV. The results are shown in Fig. 8. The effects of size on lag time shown in Fig. 8 suggest that larger tablets have a longer levodopa absorption lag time in dogs.

Table IV—Characteristics of Effervescent Enteric Tablets of Levodopa used for the Measurement of Absorption Lag Time

	Round Tablets	Oval Tablets	Sample 1	Sample 2	Sample 3
Weight per tablet, mg	594	594	369	461	474
Diameter, mm	10.6	15.7 × 8.2	9.4	10.0	10.1
Thickness, mm	6.3	5.1	5.3	5.4	5.4
Dissolution parameter, min[a]					
t_{lag}	9.0	7.4	5.1	5.1	7.0
$t_{90\text{-}10}$	1.8	1.6	0.9	1.5	2.1

[a] Dissolution parameters were determined at pH 7.5 with 20 strokes/min.

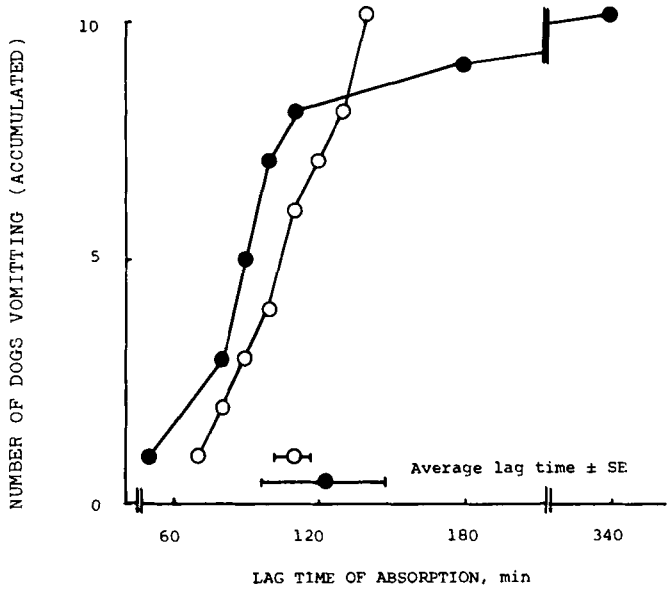

Figure 7—*Effect of tablet shape on the lag time of absorption (as indicated by emesis). Key: (○) round; (●) oval.*

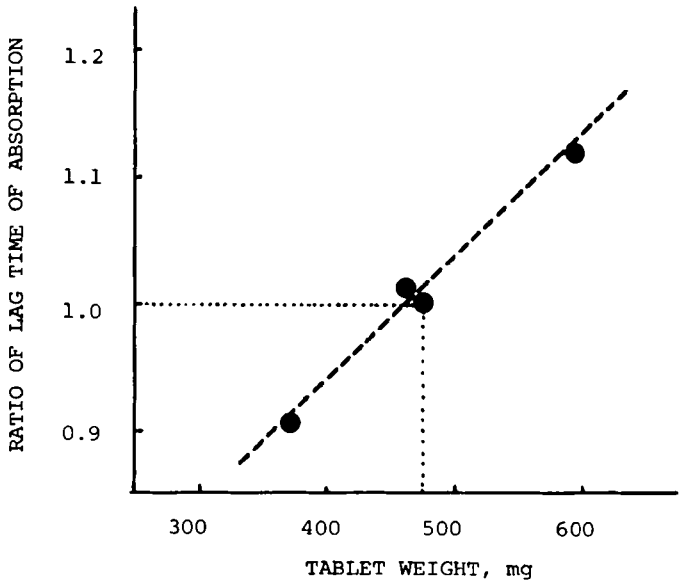

Figure 8—*Effect of tablet size on the lag time of absorption.*

CONCLUSIONS

An enteric formulation of levodopa can prevent drug absorption in the stomach and so can reduce the side effects of the drug on the stomach. Application of III-50 as an enteric coating film material resulted in an enteric tablet with rapid disintegration characteristics after passing through the stomach. Addition of an effervescent component to the tablet reduced the lag time of *in vitro* dissolution in intestinal fluid, and the rate of levodopa dissolution was accelerated. The conventional round shape was suggested as a suitable tablet shape and also a smaller size was recommended to minimize the lag time for the transit of the tablet into the intestine.

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Dissolution and Ionization of Warfarin

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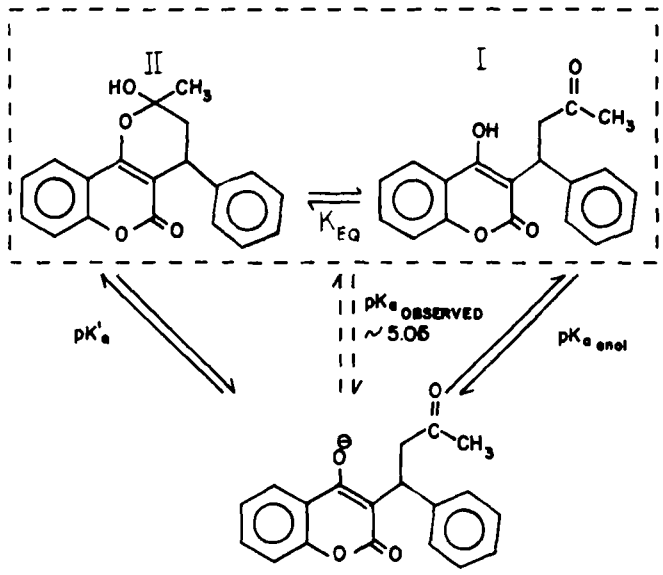
Received January 18, 1983, from the *Department of Pharmaceutical Chemistry, The University of Kansas, Lawrence, KS 66045.* Accepted for publication July 19, 1983. Present addresses: *American Cyanamid Co., Pearl River, NY 10965 and ‡Squibb Institute for Medical Research, New Brunswick, NJ 08903.

Abstract □ It has been shown in recent studies that warfarin exists in the solid state and in some nonaqueous solvents as a cyclic hemiketal. The present study was undertaken to investigate the ionization and ionization kinetics of warfarin, to confirm the probable existence of the cyclic hemiketal in aqueous solution, and to determine the possible consequences of the cyclic hemiketal to acyclic enol equilibrium and ionization kinetics on the dissolution rate of warfarin. The equilibrium aqueous solubility of un-ionized warfarin acid at 25°C and ionic strength 0.5 (with potassium chloride) was found to be 1.28×10^{-5} M, and its observed macroscopic pK_a was 5.03–5.06, depending on the method of determination. By comparing the aqueous pK_a of warfarin to phenprocoumin, a hydroxycoumarin that cannot exist in the cyclic hemiketal form, the hemiketal–acyclic enol ratio was estimated to be ~20:1. By stop-flow spectrophotometry, the ionization rate of warfarin (pH 3.5 jumped to pH 6.5) was found to have a $t_{1/2} < 1\text{–}2 \times 10^{-3}$ s. The dissolution rate of warfarin from a rotating disk (600 rpm), as a function of pH, was measured under nonbuffered but pH-stat conditions (μ = 0.5 with potassium chloride). The pH-dissolution rate profile for warfarin agreed with that calculated from an equation derived previously to describe the dissolution of instantaneous ionizing acids, *i.e.*, the profile was not perturbed from that expected from an acid of aqueous solubility 1.28×10^{-5} M (un-ionized form) and pK_a 5.06.

Keyphrases □ Dissolution—warfarin, ionization kinetics □ Warfarin—dissolution, ionization kinetics □ Ionization kinetics—warfarin, dissolution

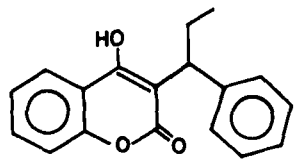
The structure of the anticoagulant warfarin is usually depicted in the open-chain form (I) whereas it is known to exist in the solid state in the cyclic hemiketal form (II) (1, 2). Spectrometric studies (3–6) have confirmed that II is also the predominant form of warfarin in solution in various nonaqueous solvents. In water, un-ionized warfarin (I and/or II) exists in equilibrium with the enolate (III) (Scheme I).

Since warfarin exists as the hemiketal in the solid state and its ionization appears to be complex, we decided to study the dissolution rate *versus* pH (unbuffered, pH maintained by pH-stat) profile of warfarin to observe whether it behaved as an instantaneously ionizing acid (7, 8). To achieve this, the dissolution rate from a compressed rotating disk of warfarin at pH 2 and in the pH range 7–9.5 was studied along with its solubility, ionization characteristics, and ionization rate. By



Scheme I

comparing the pK_a of warfarin with that of phenprocoumin (IV), a hydroxycoumarin which can exist only in an acyclic form, it was also possible to test for the existence of II as the predominant form of warfarin in aqueous solution.



IV

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