Enhanced Rectal Absorption of Theophylline, Lidocaine, Cefmetazole, and Levodopa by Several Adjuvants

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Abstract □ Ten potential adjuvants for rectal absorption which are structurally similar to salicylate have been examined using an *in situ* perfusion of the rat rectum technique as well as an *in vivo* absorption method from microenemas. All of the adjuvants studied readily disappeared from the perfusate at pH 4.5; however, several were not absorbed well at pH 7.4. Only those that were lost rapidly from the perfusate at pH 7.4 were effective in enhancing the disappearance of the drugs (theophylline, lidocaine, cefmetazole, and levodopa) at either a pH of 4.5 or 7.4. The compounds that were effective in promoting the disappearance of drugs from the rectal perfusate all had hydroxy and carboxy groups. Those substances lacking a hydroxy group were not effective. The binding of these potential adjuvants and salicylates to rat rectum tissue was studied by equilibrium dialysis. Those adjuvants with relatively high binding to rat rectal tissue were better absorbed themselves and promoted the disappearance of drugs more than those substances exhibiting little binding. Thus, adjuvant binding to some feature of the rectal membrane appears to be important in the enhanced absorption of drugs from the rectum under the conditions of this study.

Keyphrases □ Absorption, rectal—enhanced rectal absorption of theophylline, lidocaine, cefmetazole, and levodopa by several adjuvants □ Theophylline—enhanced rectal absorption by several adjuvants □ Lidocaine—enhanced rectal absorption by several adjuvants □ Cefmetazole—enhanced rectal absorption by several adjuvants □ Levodopa—enhanced rectal absorption by several adjuvants

In other papers (1–3), salicylate has been shown to significantly improve the absorption of theophylline, lidocaine, cefmetazole, and levodopa from the rectum, particularly in their ionic forms. The present report describes the effects of several other compounds which are structurally similar to salicylate on the rectal absorption of these drugs. These potential adjuvants include the sodium salts of benzoic acid, *o*-anisic acid, *p*-anisic acid, 3-methoxysalicylic acid, 5-methoxysalicylic acid, 2,4-dihydroxybenzoic acid, 2,5-dihydroxybenzoic acid, 3,5-dihydroxybenzoic acid, 2,4-dimethoxybenzoic acid, and homovanillic acid. The drugs chosen for this study represent several different classes: theophylline is a neutral substance, lidocaine is a base, cefmetazole is an acid, and levodopa exists as a zwitterion in solution.

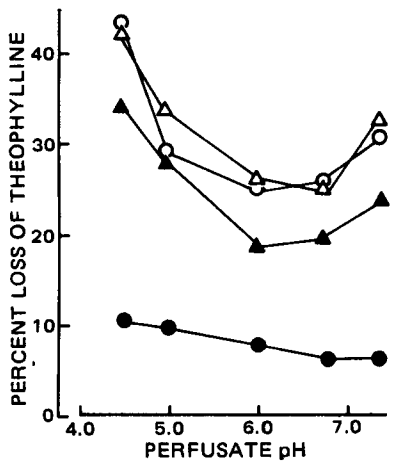

Figure 1—*The percent loss of theophylline from a perfusate of the rat rectum after 1 hr as a function of pH in the presence of 0.5% sodium homovanillate (○); sodium 5-methoxysalicylate (△); sodium 2,4-dihydroxybenzoate (▲); without an adjuvant (●).*

EXPERIMENTAL

Materials—Theophylline[1], sodium cefmetazole[2], and levodopa[2] were used as obtained from the manufacturer. Salicylic acid[3], benzoic acid[3], *o*-anisic acid[3], *p*-anisic acid[3], 3-methoxysalicylic acid[3], 5-methoxysalicylic acid[3], 2,4-dihydroxybenzoic acid[3], 2,5-dihydroxybenzoic acid[3], 3,5-dihydroxybenzoic acid[3], 2,4-dimethoxybenzoic acid[3], and homovanillic acid[3] were converted to the sodium salt by reacting the acid form with either sodium bicarbonate in water followed by recrystallization from ethanol or by reacting the acid with sodium ethoxide in anhydrous ethanol while bubbling nitrogen through the solution. Lidocaine[1] was converted to the hydrochloride salt by mixing with an equimolar solution of hydrochloric acid and evaporating to dryness without heat.

Animals—Sprague-Dawley male rates (200–300 g) were fasted for 16 hr prior to the experiments. During the experiment, the rats were kept on a 38° surface and were anesthetized with pentobarbital (60 mg/kg).

The *in situ* perfusion studies using the rat rectum and the *in vivo* absorption studies were carried out as described previously (2) with an ionic strength of 0.75. The pH of the perfusate was maintained constant by either the addition of 0.1 *N* NaOH or 0.1 *N* HCl, as needed. The analysis of theophylline, lidocaine, cefmetazole, and levodopa were performed by HPLC as described previously (2, 3). The analyses of the adjuvants used in the present study were done using HPLC with a reversed-phase column, 25 cm in length, a flow rate of 1.0 ml/min, and detection at 254 nm. The mobile phases consisted of mixtures of 0.1 *M* acetate buffer–methanol with ratios: 30:70 for benzoate, 2,4-dimethoxybenzoate, *o*-anisate, and *p*-anisate; 60:40 for homovanillate; 3-methoxysalicylate, and 5-methoxysalicylate; and 20:80 for 2,4-dihydroxybenzoate, 2,5-dihy-

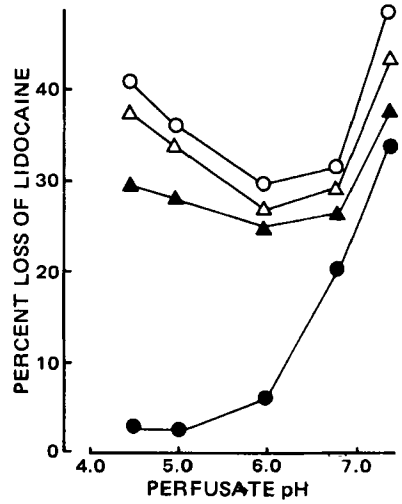

Figure 2—*The percent loss of lidocaine from a perfusate of the rat rectum after 1 hr as a function of pH in the presence of 0.5% sodium homovanillate (○); sodium 5-methoxysalicylate (△); sodium 2,4-dihydroxybenzoate (▲); without an adjuvant (●).*

[1] Sigma Chemical Co.
[2] Sankyo.
[3] Aldrich Chemical Co.

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Table I—The Percent Loss of Drug and Adjuvant from Perfusate after 60 min at pH 4.5 [a]

Adjuvant	Theophylline	Lidocaine	Levodopa	Cefmetazole	Adjuvant[b]	Adjuvant[c]
—	10.3 ± 1.2	2.8 ± 0.9	2.4 ± 0.3	19.1 ± 2.8		
Benzoate	8.6 ± 1.2	—	2.3 ± 0.9	16.5 ± 5.2	40.6 ± 2.9	38.3 ± 3.2
o-Anisate	9.2 ± 0.7	—	3.5 ± 0.9	17.7 ± 3.9	36.3 ± 4.8	37.2 ± 5.6
p-Anisate	8.9 ± 0.7	—	2.1 ± 1.1	19.2 ± 1.9	40.2 ± 5.3	39.4 ± 2.8
3-Methoxysalicylate	34.5 ± 2.9[d]	40.6 ± 4.2[d]	28.4 ± 3.3[d]	39.6 ± 3.2[d]	30.3 ± 2.3	31.2 ± 3.5
5-Methoxysalicylate	42.6 ± 6.2[d]	37.5 ± 8.7[d]	30.4 ± 3.8[d]	51.6 ± 3.8[d]	32.5 ± 5.6	31.8 ± 4.3
2,4-Dihydroxybenzoate	34.3 ± 7.2[d]	29.5 ± 7.8[d]	22.5 ± 6.2[d]	39.6 ± 4.3[d]	29.8 ± 7.2	27.2 ± 2.5
2,5-Dihydroxybenzoate	29.8 ± 4.0[d]	34.7 ± 4.1[d]	23.8 ± 5.3[d]	32.6 ± 3.9[d]	26.3 ± 4.3	24.6 ± 3.2
3,5-Dihydroxybenzoate	25.2 ± 9.0[d]	24.9 ± 3.4[d]	18.6 ± 3.1[d]	28.4 ± 2.5[d]	20.5 ± 2.8	19.5 ± 3.2
2,4-Dimethoxybenzoate	7.9 ± 2.3	—	1.9 ± 0.8	15.7 ± 4.1	27.4 ± 4.8	27.7 ± 2.8
Homovanilate	43.5 ± 5.8[d]	40.8 ± 6.9[d]	28.5 ± 7.4[d]	43.5 ± 8.1[d]	30.5 ± 4.4	28.8 ± 2.6

[a] The uncertainties represent standard deviations. [b] Initial concentration of adjuvant was 0.5%. [c] Initial concentration of adjuvant was 2%. [d] $p < 0.001$ when compared to no adjuvant using a standard t test ($n \geq 4$).

Table II—The Percent Loss of Drug and Adjuvant from Perfusate after 60 min at pH 7.4 [a]

Adjuvant	Theophylline	Lidocaine	Levodopa	Cefmetazole	Adjuvant[b]	Adjuvant[c]
—	6.1 ± 0.8	33.5 ± 7.8	3.2 ± 1.1	2.6 ± 1.8		
Benzoate	5.2 ± 1.1		2.9 ± 0.9	3.1 ± 1.2	2.2 ± 1.5	
o-Anisate	4.1 ± 1.7		3.5 ± 0.7	2.8 ± 0.8	3.7 ± 1.5	
p-Anisate	6.1 ± 2.4		3.4 ± 1.7	3.5 ± 1.6	2.9 ± 1.3	
3-Methoxysalicylate	30.8 ± 5.3[d]		23.6 ± 3.5[d]	31.4 ± 4.3[d]	23.6 ± 4.2	14.1 ± 2.8
5-Methoxysalicylate	32.8 ± 9.3	43.5 ± 9.2	25.8 ± 4.3[d]	37.1 ± 7.7[d]	24.3 ± 6.2	14.7 ± 3.2
2,4-Dihydroxybenzoate	23.8 ± 7.1	37.8 ± 9.3	21.5 ± 6.3[d]	24.9 ± 6.7	18.4 ± 7.3	8.7 ± 2.8
2,5-Dihydroxybenzoate						
3,5-Dihydroxybenzoate	17.8 ± 2.1[d]		18.6 ± 3.1[d]	18.3 ± 1.8[d]	10.5 ± 4.8	6.1 ± 1.6
2,4-Dimethoxybenzoate	6.8 ± 1.9		5.2 ± 3.1	2.9 ± 1.8	6.4 ± 2.8	
Homovanilate	30.7 ± 5.2	48.3 ± 8.4	23.8 ± 6.2[d]	31.8 ± 8.6[d]	21.5 ± 3.8	11.8 ± 2.8

[a] The uncertainties represent standard deviations. [b] Initial adjuvant concentration was 0.5%. [c] Initial adjuvant concentration was 2%. [d] $p < 0.001$ when compared to no adjuvant using a standard t test.

droxybenzoate, and 3,5-dihydroxybenzoate. The absolute bioavailabilities were determined as described previously (2).

Binding Study of Adjuvants to Rat Rectal Tissue—The binding of various adjuvants with rat rectal tissue was studied using an equilibrium dialysis method. Dialysis tubing containing 1.4 mg of adjuvant and 200 mg of rectal tissue in 2.0 ml of 0.0667 *M* phosphate buffer at pH 7.4 was suspended in a 10-ml test tube containing 5 ml of the 0.0667 *M* phosphate buffer used earlier. After equilibration for 48 hr at 4°, the concentration of adjuvant in the outside solution was measured. The percent binding of adjuvant to 200 mg of rectal tissue was calculated from the following relationship:

$$\text{Percent Binding} = \{[T - (CV)]/T\}100 \qquad \text{(Eq. 1)}$$

where T, C, and V represent the total amount of adjuvant, the concentration of adjuvant in the outer solution, and the total volume of buffer (7.0 ml), respectively. The amount of adjuvant that is not bound is equal to CV.

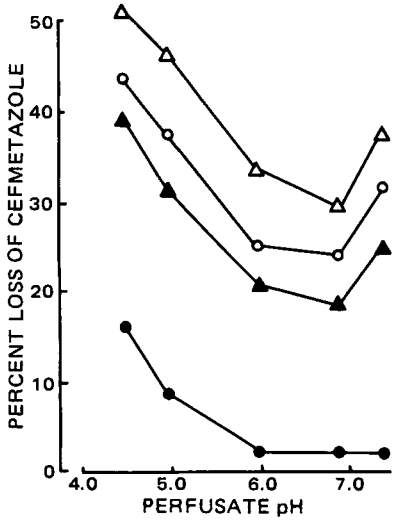

Figure 3—*The percent loss of cefmetazole from a perfusate of the rat rectum after 1 hr as a function of pH in the presence of 0.5% sodium homovanillate (○); sodium 5-methoxysalicylate (△); sodium 2,4-dihydroxybenzoate (▲); without an adjuvant (●).*

RESULTS AND DISCUSSION

Effect of Adjuvants on Drug Disappearance from Perfusate—Ten compounds have been studied regarding their effectiveness in enhancing the loss of theophylline, lidocaine, cefmetazole, and levodopa from a perfusing solution in the rat rectum. The pH profiles of the effects of three of the most potent absorption promoters, *i.e.*, homovanillate, 5-methoxysalicylate, and 3,5-dihydroxybenzoate, on the disappearance of theophylline, lidocaine, cefmetazole, and levodopa from the perfusate after 1 hr are shown in Figs. 1–4.

The disappearance of each of the drugs from the perfusing solution was significantly facilitated by the presence of each of these three adjuvants. The enhancement of loss from the perfusate was particularly evident at pH values >7.4 and <5.0. As shown in Fig. 5, the loss of adjuvant from the perfusate paralleled the extent of disappearance of the drugs studied. Similar results were observed using sodium salicylate as an adjuvant (2). Furthermore, as shown in Fig. 6, the disappearance of these three adjuvants was not affected by the presence or absence of the four drugs studied in the perfusate. This indicates that the enhancement of drug absorption by these adjuvants is not due to the formation of a complex with the drugs.

The effects of the 10 adjuvants studied on the disappearance of theophylline, lidocaine, cefmetazole, and levodopa from a rectal perfusate are summarized in Tables I and II for pH 4.5 and 7.4, respectively. The

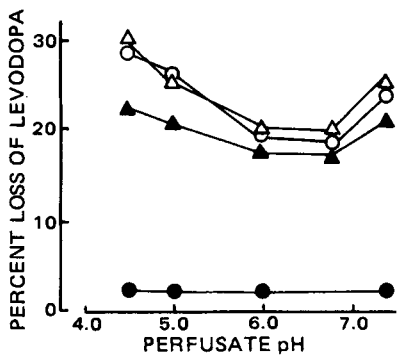

Figure 4—*The percent loss of levodopa from a perfusate of the rat rectum after 1 hr as a function of pH in the presence of 0.5% sodium homovanillate (○); sodium 5-methoxysalicylate (△); sodium 2,4-dihydroxybenzoate (▲); without an adjuvant (●).*

Table III—Absolute Bioavailabilities of a 15-mg/kg Dose of Theophylline and Cefmetazole [a]

Dose of 5-Methoxysalicylate, mg/kg	Bioavailability, % Theophylline	*n*	Cefmetazole	*n*
0	32.5 ± 12.4	4	7.8 ± 1.3	4
10	83.5 ± 9.2	1	18.5	1
15	128.4 ± 32.5	4	38.3 ± 4.2	4
30			48.6 ± 7.2	4
50			76.8	1
Dose of Sodium Homovanillate				
0	32.5 ± 2.4	4	7.8 ± 1.3	4
7.5	69.5 ± 14.8	4		
15	119.7 ± 28.5	4	21.8	1
30			34.7 ± 5.4	4
50			51.2 ± 8.4	4

[a] After rectal administration as a microenema in the presence of 5-methoxysalicylate and homovanillate.

extent of the disappearance of the adjuvants themselves is also shown.

All of the adjuvants studied readily disappeared from the perfusate at pH 4.5. However, at pH 7.4 the disappearance of all the adjuvants was somewhat lower and very low for benzoate, *o*- and *p*-anisate, and 2,4-dimethoxybenzoate. Those adjuvants that were not well absorbed at pH 7.4 did not significantly enhance the disappearance of the drugs at pH 4.5 or 7.4. However, those compounds, which readily disappeared from the perfusate at pH 7.4, facilitated the loss of drugs from the perfusates at pH 4.5 and 7.4.

The disappearance of the adjuvants from the perfusate as reflected by the percent lost after 1 hr did not depend on the initial concentration at pH 4.5. However, the loss of adjuvant at pH 7.4 did depend on the initial concentration with a greater percent loss occurring at the lower concentrations, as shown in the last two columns of Table II.

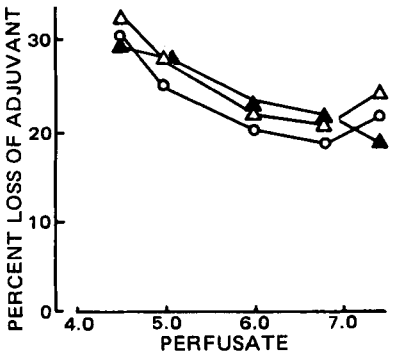

Figure 5—*The percent loss of sodium homovanillate (○), 5-methoxysalicylate (△) and 2,4-dihydroxybenzoate (▲) from a perfusate of the rat rectum after 1 hr as a function of pH. Initial concentration is 0.5%.*

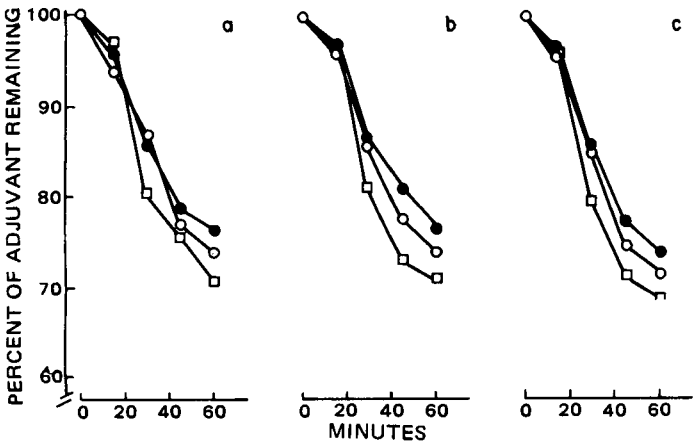

Figure 6—*Percent of homovanillate (●), 5-methoxysalicylate (○), and 2,4-dihydroxybenzoate (□) remaining in the perfusate as a function of time in the presence of (a) theophylline, (b) cefmetazole, or (c) in the absence of drug.*

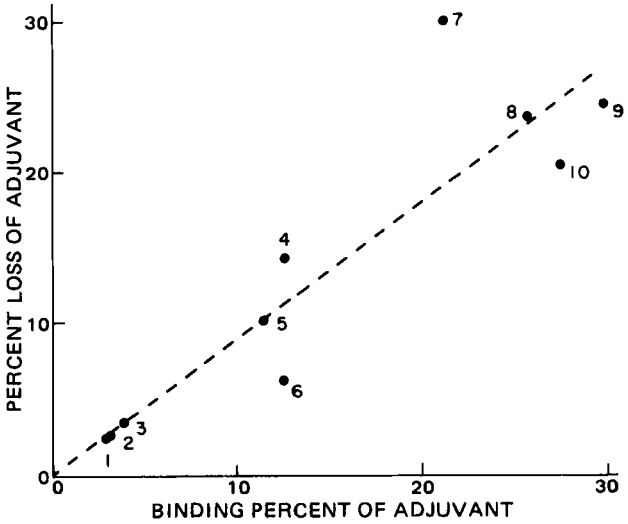

Figure 7—*Percent loss of adjuvant from perfusate at 60 min* versus *the percent binding of adjuvant to rat rectal tissue. Key: 1, benzoate; 2, p-anisate; 3, o-anisate; 4, 2,4-dihydroxybenzoate; 5, 3,5-dihydroxybenzoate; 6, 2,5-dihydroxybenzoate; 7, salicylate; 8, 3-methoxysalicylate; 9, 5-methoxysalicylate; 10, homovanillate.*

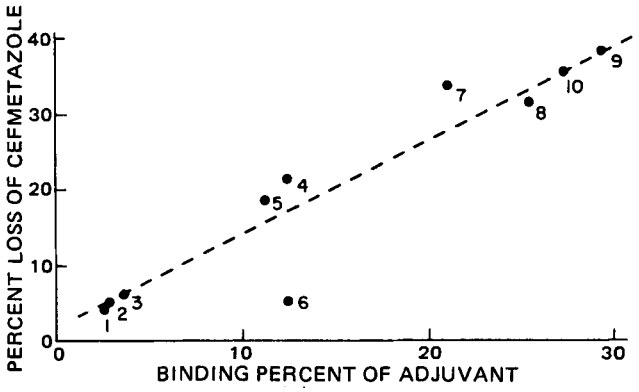

Figure 8—*Percent loss of cefmetazole from perfusate at 60 min* versus *the percent binding of adjuvant to rat rectal tissue. Key: 1, benzoate; 2, p-anisate; 3, o-anisate; 4, 2,4-dihydroxybenzoate; 5, 3,5-dihydroxybenzoate; 6, 2,5-dihydroxybenzoate; 7, salicylate; 8, 3-methoxysalicylate; 9, 5-methoxysalicylate; 10, homovanillate.*

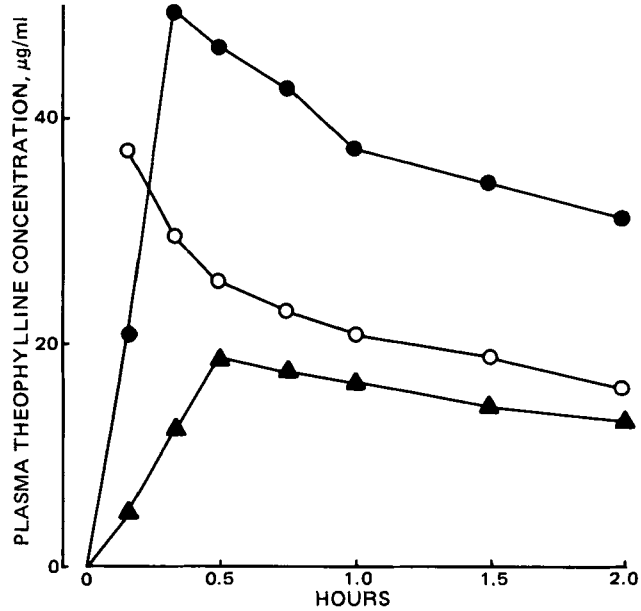

Figure 9—*Theophylline concentration (μg/ml) from an intravenous injection of 10 mg/kg (○) and from a microenema containing 15 mg/kg of theophylline (▲,●) and 15 mg/kg of sodium homovanillate (●).*

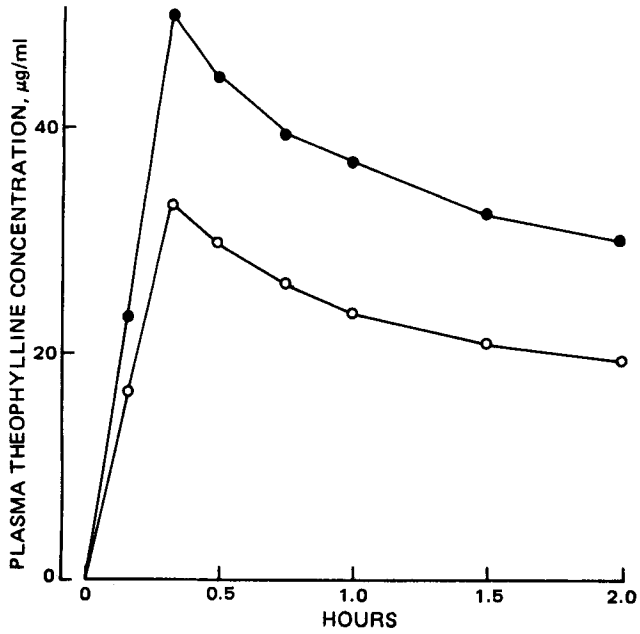

Figure 10—*Plasma theophylline concentration (μg/ml) from a microenema containing 15 mg/kg of theophylline and either 7.5 mg/kg (○) or 15 mg/kg (●) of 5-methoxysalicylate.*

The compounds that were effective in promoting the disappearance of drugs from the rectal perfusate all have hydroxy and carboxy groups. Benzoate, *o*- and *p*-anisate, and 3,5-dimethoxybenzoate do not have hydroxy groups, did not facilitate the disappearance of drugs from the perfusate, and did not disappear from the perfusate themselves at pH 7.4. It appears that for those substances which are chemically similar to salicylate, the presence of both the hydroxy and carboxy groups is important.

Binding of Adjuvants with Rat Rectum Tissue—The facts that those compounds which act as adjuvants for rectal absorption, both in this study and previous studies, disappear from the rectal perfusate even in their ionic form and that disappearance at pH 7.4 depends on their initial concentrations suggests that their potentiating effect may depend on their binding to some feature of the rat rectal tissue. Therefore, a binding study of these potential adjuvants and salicylate with rat rectal tissue was conducted at 4° using equilibrium dialysis.

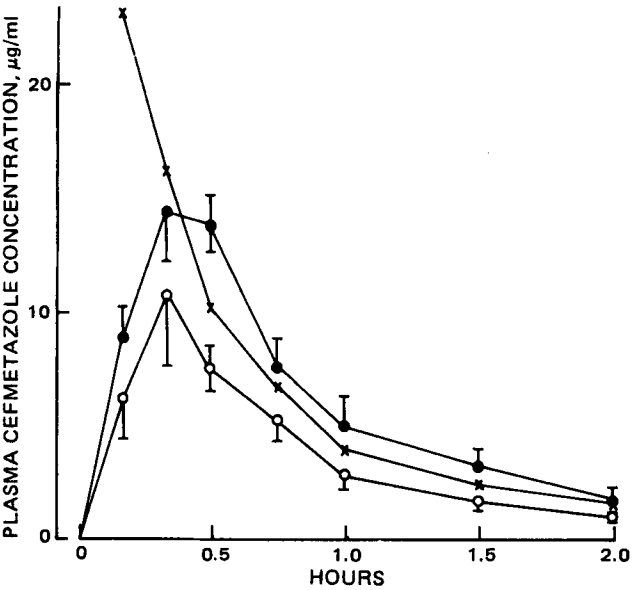

Figure 11—*Plasma cefmetazole concentration (μg/ml) in the rat from an intravenous injection of 10 mg/kg (x) and from a microenema containing 30 mg/kg of cefmetazole (○,●) and 30 mg/kg (○) or 50 mg/kg (●) of sodium homovanillate.*

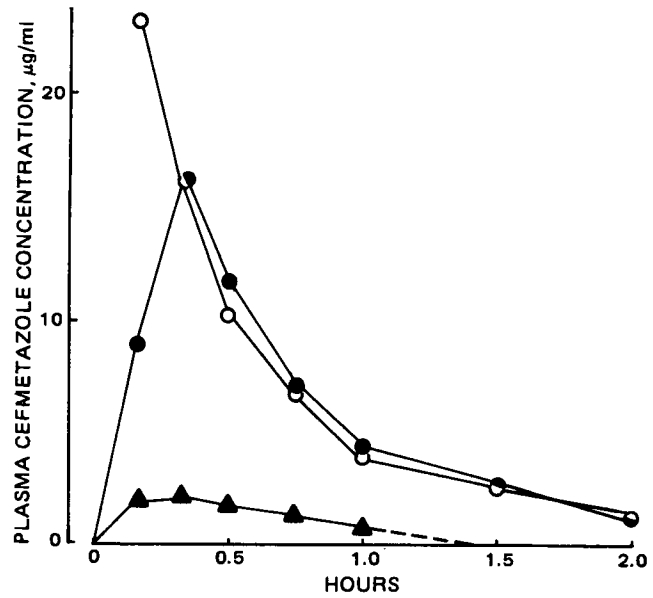

Figure 12—*Plasma cefmetazole concentration (μg/ml) in the rat from an intravenous injection of 10 mg/kg (○) and from a microenema containing 30 mg/kg cefmetazole (▲,●) and 30 mg/kg (●) of 5-methoxysalicylate or no adjuvant (▲).*

The relationship between the percent loss of adjuvant from perfusate at pH 7.4 and the extent of binding of the adjuvant with fat rectal tissue at the same pH is shown in Fig. 7. Figure 8 compares the percent loss of cefmetazole after a 1-hr perfusion in the presence of adjuvant with the percent of binding of the adjuvant. These data show a good correlation between the amount of adjuvant and drug lost and the binding of the adjuvant to rectal tissue. Those adjuvants with relatively high binding to the rectal tissue disappeared to a greater extent and promoted the disappearance of drugs more than those with little binding.

Thus, it appears that binding to some feature of the rectal membrane is necessary for facilitated rectal absorption of drugs and may suggest some type of active transport.

Effects of Adjuvants on Plasma Levels—The effect of homovanillate and 5-methoxysalicylate on the absorption of theophylline and cefmetazole after rectal administration as a microenema at pH 7.4 as reflected in plasma levels of the drug are shown in Figs. 9–12. In each case the absorption was rapid and facilitated by the presence of either homovanillate or 5-methoxysalicylate in the microenema solution and depended on the concentration of the adjuvant. Absolute bioavailabilities of theophylline and cefmetazole as a function of the concentration of homovanillate and 5-methoxysalicylate are shown in Table III.

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(1) T. Nishihata, J. H. Rytting, and T. Higuchi, *J. Pharm. Sci.*, **69**, 744 (1980).

(2) *Ibid.*, **70**, 71 (1981).

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