Enhanced Rectal Bioavailability of Polypeptides Using Sodium 5-Methoxysalicylate as an Absorption Promoter

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Abstract □ The absorption-promoting effect of sodium 5-methoxysalicylate was studied in the rat with respect to rectal delivery of pentagastrin and gastrin. Rectal bioavailability was quantitated by direct comparison of pharmacological effect with intravenous dose response. Coadministration of the absorption adjuvant greatly enhanced the rectal bioavailability of the model polypeptides. Sodium 5-methoxysalicylate, therefore, is representative of a new type of absorption promoter which appears to facilitate rectal absorption of polypeptide drug entities.

Keyphrases □ Bioavailability—rectal, absorption of small polypeptides, rats □ Polypeptides—pentagastrin and gastrin, rectal bioavailability, rats □ Adjuvant—absorption effect of sodium 5-methoxysalicylate, rats

Parenteral injection has long been the only reliable method to administer polypeptide drugs. Successful oral dosage forms of biologically active polypeptides have traditionally been elusive (1). However, there has been some limited success using the rectal delivery route as an alternative to the parenteral route.

Surface-active agents have been shown to increase rectal absorption of water-soluble polypeptides such as insulin (2). More recently, enhanced rectal delivery of theophylline and lidocaine using sodium salicylate as an absorption promoter was reported (3, 4), and enhanced rectal delivery of insulin in rats using 5-methoxysalicylate as an absorption adjuvant has been demonstrated (5). The use of salicylates as rectal absorption adjuvants represents a divergence from the traditional surfactant-enhanced delivery of poorly absorbed drugs. Furthermore, nontoxic salt forms of salicylate appear to be less tissue damaging than surfactant-type absorption adjuvants (3, 6).

The present study was designed to evaluate the feasibility of rectal delivery of small polypeptides using 5-methoxysalicylate as an absorption promoter.

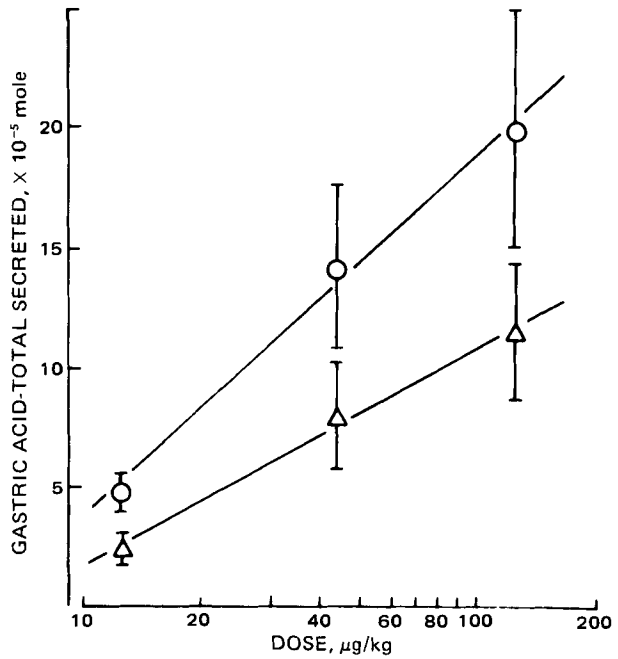

Figure 1—*Intravenous dose response of total gastric acid secretion following intravenous administration of gastrin (△) and pentagastrin (○) to anesthetized rats. The drug dosages were 12.5, 44.0, and 125 μg/kg. The error bars represent standard deviations with n=6.*

EXPERIMENTAL

Male Long-Evans rats, 230–290 g, were fasted 36 hr prior to experimentation. Rats were anesthetized with pentobarbital sodium (50 mg/kg ip) and surgically prepared for gastric perfusion according to the method of Ghosh and Schild (7). The stomachs were rinsed and ultimately perfused with 20 ml of physiological saline solution. The perfusion rate was 2 ml/min provided by a peristaltic pump. Perfusate and rat body temperature were held constant at 38° and the pH of the perfusate was held constant at 4.0[1] with 0.05 *N* NaOH as the titrant. Stomach acid output was continuously monitored by recording the amount of titrant required to maintain a constant pH with respect to time.

The target polypeptides for rectal delivery were pentagastrin[2] (5 residues) and gastrin[3] (17 residues). Aqueous polypeptide solutions were made up and adjusted to pH 8.0 using ammonium hydroxide and the ionic strength was adjusted to $\mu = 0.15$ with sodium chloride. Polypeptide concentration in these solutions was 250 μg/ml. Both pentagastrin and gastrin were administered intravenously (external jugular) to obtain dose–response curves.

Identical drug solutions were also administered rectally as microenema formulations (0.5 ml/kg) with or without the addition of 50 mg/ml of

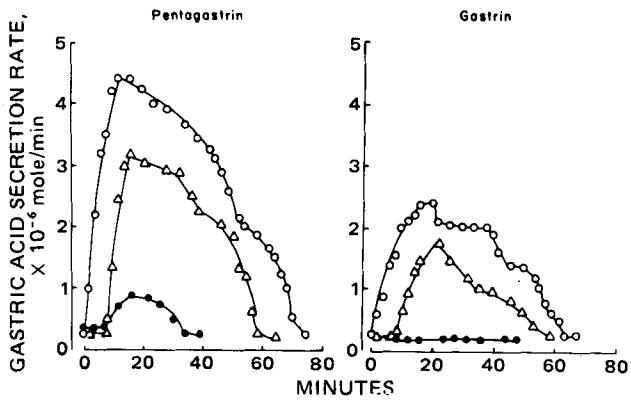

Figure 2—*Typical, single animal profiles of gastric acid secretion following administration of pentagastrin and gastrin to anesthetized rats. The solutions were administered: intravenous with 125 μg polypeptide/kg in saline (○); rectally with 125 μg polypeptide/kg in saline, pH 8.0 (●); and rectally with 125 μg polypeptide/kg rat and 25 mg 5-methoxysalicylate/kg rat in saline, pH 8.0 (△).*

[1] pH Stat, Radiometer, Copenhagen.
[2] Peptavalon, Ayerst Laboratories, Inc.
[3] Porcine intrail mucosa, Sigma. A mixture of gastrin I and gastrin II prepared through Sephadex stage as described by Gregory and Tracy (8).

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Table I—Total Amount of Gastric Secretion Stimulated by Pentagastrin

Experiment No.	Gastric Acid Secreted, $\times 10^{-5}$ mole				
	Intravenous Administration			Microenema 125 μg/kg[a]	
	12.5[a]	44[a]	125[a]	No Adjuvant	Adjuvant[b]
1	3.3	10.0	22.6	2.9	21.4
2	4.8	9.1	15.9	0.6	12.5
3	6.0	14.0	19.2	2.3	10.6
4	5.5	15.9	29.6	1.0	11.3
5	4.2	17.0	18.0	0.2	10.4
6	5.0	18.8	13.5	2.7	10.3
Mean	4.8	14.1	19.8	1.6	12.9
SD	0.9	3.5	5.2	1.1	3.9
Pharmacologic effect Relative to Intravenous, %			100	8	65
Bioavailability[c], %			100 (defined)	6	33

[a] Dose of pentagastrin (μg/kg). [b] Sodium 5-methoxysalicylate (25 mg/kg). [c] Based on dose–response curve, effect *versus* log dose.

Table II—Total Amount of Gastric Secretion Stimulated by Gastrin

Experiment Number	Gastric Acid Secreted, $\times 10^{-5}$ mole				
	Intravenous Administration			Microenema, 125 μg/kg[a]	
	12.5[a]	44[a]	125[a]	No Adjuvant	Adjuvant[b]
1	3.8	6.4	7.6	0	4.6
2	2.8	5.6	14.1	0	1.6
3	1.7	11.3	8.9	0	7.3
4	2.1	8.6	14.6	0	4.6
5	1.4	9.5	10.4	0	4.4
6	2.4	5.3	12.2	0	6.9
Mean	2.4	7.8	11.3	0	4.9
SD	0.8	2.2	2.6		1.9
Pharmacologic effect Relative to intravenous, %			100	0.0	43
Bioavailability[c], %			100 (defined)	—	18

[a] Dose of gastrin (μg/kg). [b] Sodium 5-methoxysalicylate (25 mg/kg). [c] Based on dose response curve, effect *versus* log dose.

sodium 5-methoxysalicylate. After the addition of the sodium 5-methoxysalicylate as a potential absorption adjuvant, the microenema was readjusted to pH 8.0 before administration. As a separate control, a solution of 5-methoxysalicylate without the polypeptides was administered.

RESULTS AND DISCUSSION

Gastrin-like compounds were chosen for this study for several reasons. Gastric acid secretion is relatively easy to monitor in a test animal and at the same time is particularly sensitive to circulating hormone levels. The relationship between pharmacological response and the logarithm of the intravenous dose is relatively linear over a significant dosage range (Fig. 1)[4]. This allows for a reasonable bioavailability assessment of the rectally administered aqueous polypeptide solutions by comparing the pharmacological effect with the intravenous dose–response curve.

Figure 2 shows typical, single animal plots of gastric acid secretion rate after intravenous or rectal administration of both pentagastrin and gastrin at doses of 125 μg/kg. The area under the curve represents the total amount of gastric acid secretion in response to administration of either pentagastrin or gastrin in the presence or absence of adjuvant. The time course of the pharmacological response following intravenous administration did not decline in an apparent zero-order fashion and, therefore, may not directly represent the logarithm of the drug–plasma concentration. Nevertheless, the response profiles are similar regardless of administration route and should allow for a reasonable rectal bioavailability assessment based on the intravenous dose response.

Rectal administration of 125 μg/kg of pentagastrin resulted in only 6 ± 4% bioavailability relative to intravenous dose response (uncertainties are expressed as standard deviations) (Table I). The addition of 5 mg of sodium 5-methoxysalicylate to the microenema resulted in a bioavailability of 33 ± 10%, a five-fold increase in drug delivery to the active site. The rectal administration of 125 μg/kg of gastrin without the absorption adjuvant resulted in no detecetable response (Table II). However, the addition of the absorption adjuvant to the microenema resulted in 18 ± 7% drug delivery.

In a separate series of experiments, the concentration of absorption adjuvant in the microenema formulations was lowered from 5 to 1% (w/v). In these experiments the bioavailability of rectally administered gastrin was only 8%, suggesting a dose-response relationship between adjuvant concentration and the absorption promoting effect.

The microenema formulations containing 5% sodium 5-methoxysalicylate are hyperosmotic. In control experiments in which an osmolar equivalent of sodium chloride was substituted for the adjuvant, no enhancement of rectal absorption was seen due to an osmolarity effect. Additional control experiments in which 5-methoxysalicylate was administered rectally without the polypeptide hormone produced no change in stomach acid output. Thus, the observed improvement in rectal polypeptide delivery must be attributed to the adjuvant, which appears to effect a temporary change in the normal mucosal permeability of the rectal compartment.

From this study, it is apparent that sodium 5-methoxysalicylate is representative of a new type of absorption promoter which facilitates nonparenteral polypeptide drug delivery. As nonparenteral alternatives, rectal dosage forms such as microenemas and suppositories may offer a convenient means of expanding polypeptide drug therapy beyond the clinical setting.

[4] The response to higher dosage appears to be more variable than the response in the lower dosage range.

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