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The Effect of Detergent Treatment of the Gastric Mucosa on Drug Transport (39295)

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(Introduced by G. E. Cartwright)

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It is well known that following systemic administration drugs and other foreign compounds may be recovered from gastric juice (1, 2). Recovery of drugs from the stomach has therefore been utilized as a means of drug disposal in the treatment of acute drug intoxications (2, 3). For a variety of reasons, however, many drugs do not readily diffuse into gastric juice and this route of disposal is ineffectual. Accordingly, it would be desirable to develop techniques which facilitate or enhance transport of these compounds, as well as those known to enter gastric juice. Since the permeability characteristics of biologic membranes has been shown to be altered by treatment with surface acting agents (4–8) we have treated the gastric mucosa of dogs with a detergent in order to explore its effects on the transport of drugs from the systemic circulation to gastric juice.

Methods and materials. Following an overnight fast, mongrel dogs weighing approximately 20 kg were prepared for laparotomy. The dogs were anesthetized with intravenous sodium pentabarbital, underwent endotrachial intubation, and had their ventilation controlled with the Harvard animal respirator. Arterial blood pressure was continuously measured and recorded on a Sanborn model 7714 polygraph via a catheter placed in the femoral artery which was attached to a Statham strain gauge. Normal saline and blood were infused to maintain blood pressure via catheters placed in the femoral veins. Test drugs were infused through these intravenous catheters.

The stomach was prepared according to the Chamber technique of Moody *et al.* (9). This technique involves isolation of the greater curvature of the body of the stomach and its blood supply which is via a branch of the splenic artery and application of a circular lucite chamber (diameter, 6 cm; capacity, 17 cm^2/hemichamber). The chamber is equally divided into two watertight compartments by means of a plastic strut. The chambers are filled through an injection port at the top of the chamber and drained by gravity through emptying ports at the bottom. This is facilitated by mounting the chamber over the anterior abdominal wall at an angle of 30°. The amount of fluid instilled and recovered is determined by weight to 0.01 g on a Mettler balance. This technique enables the simultaneous measurement of recovered drugs and secretions from both chambers; one may therefore be used as a test side and the other as the control. Previous studies have documented the blood flow, oxidative metabolism, and permeability of this preparation under a variety of conditions (10–12).

Drug transport was studied with the canine stomach in a basal, nonstimulated state. At the beginning of each experiment both chambers were filled with approximately 15 ml of isotonic 0.15 *N* HCl. Following a 15-min equilibration period the chambers were drained. Six 15-min baseline periods were obtained. A test side was then randomly selected and treated with 1% (35 m*M*) sodium lauryl sulfate made isotonic with hydrochloric acid for two consecutive 15-min periods (7, 8) while the control side received the standard 0.15 *N* HCl solution. Thereafter, both sides received 0.15 *N* HCl until the last period of the experiment (13) when the control side was treated with sodium lauryl sulfate.

Transport of two compounds, antipyrine (pK_a 1.4) and aminopyrine (pK_a 5.0) was studied. Each dog was given a loading dose of aminopyrine (20 mg/kg) and antipyrine (60 mg/kg); a constant infusion of aminopyrine (5 mg/kg/hr) and antipyrine (6 mg/kg/hr) was provided with a Harvard infusion pump.

Instilled and recovered solutions were analyzed for sodium by flame photometry (Radiometer). Antipyrine was measured according to Brody *et al.* (13). Aminopyrine was determined according to the method of Brody and Axelrod (14).

The potential difference (PD) across the gastric mucosa was measured for each period. This was accomplished by placing a reference electrode in the femoral vein and an exploring electrode on the gastric mucosa through the chamber port. The electrodes consisted of polyvinyl catheters with an internal diameter of $^1/_{16}$-in. filled with saturated KCl in 3% agar. The electrodes were connected to a Beckman pHASAR pH meter by means of balanced Calomel electrode in saturated KCl solution.

Back diffusion was determined by titrating instilled and recovered isotonic HCl solutions to pH 7 (radiometer titrator).

Hemoglobin determinations on the recovered test and control solutions were made using a cyanmethemoglobin method (Hycel). Statistical significance of the data was determined by analysis with the paired *t* test.

Results. Antipyrine recovered from the instilled fluid in eight experiments is presented in Table I. Since we compared hemichambers in our preparation we did not attempt to obtain a steady state for the administered drugs. This is reflected by an increase of drug recovery during progress of the experiments. Mean basal, pretreatment values (periods 1-6) varied between dogs and from one side of the chamber to the other, presumably reflecting differences in gastric mass and/or transport capacity. Following detergent treatment (periods 7-8) antipyrine recovery increased substantially over the control side: mean cumulative antipyrine recovery for periods 7-12 on the control sides was 2142 μg; whereas it was 3261 μg on the sides treated with detergent.

To eliminate hemichamber differences in capacity of the gastric mucosa to transport drugs, basal drug recovery for periods 1-6 were averaged and assigned a value of 100%. Drug recovery during treatment periods was thereafter expressed as a percentage of the basal. Figure 1 compares the results of antipyrine recovery for the treated and untreated sides of all experiments ex-

TABLE I. RECOVERY OF ANTIPYRINE (μg/15 min) FROM GASTRIC HEMICHAMBERS.

Experiment	Hemichamber	Basal (periods 1-6)		Treatment periods							
		Mean	Total	7[a]	8[a]	9	10	11	12	Total	13[b]
1	Test	247	1474	562	541	583	578	404	494	3162	145
	Control	202	1210	164	273	222	175	153	193	1180	481
2	Test	228	1366	525	712	—[d]	708	458	673	3075	496
	Control	254	1525	498	517	—[d]	489	482	615	2606	627
3	Test	163	978	416	408	434	508	538	529	3528	350
	Control	151	908	249	269	312	299	323	439	1941	578
4	Test	202	1211	382	444	445	452	474	574	2772	595
	Control	211	1263	237	261	260	268	307	365	1694	764
5	Test	265	1327	670	654	751	—[c]	—	—	2075[e]	—
	Control	259	1295	308	309	282	—[c]	—	—	899	—
6	Test	528	3169	694	714	802	750	690	676	4326	669
	Control	544	3267	533	532	549	521	521	549	3205	711
7	Test	276	1659	454	587	366	440	396	397	2640	374
	Control	309	1857	319	302	299	275	276	323	1794	441
8	Test	611	3667	757	804	782	728	704	735	4510	718
	Control	599	3596	643	643	643	618	641	629	3817	753
									582		
Mean	Test	315	1856	557	608	594	594	523	444	3261	478
	Control	316	1865	368	388	366	377	386		2142	622

[a] Detergent applied to test hemichamber.
[b] Detergent applied to control hemichamber.
[c] Experimental design altered at period 10.
[d] Samples lost.
[e] Totals for periods 7-9.

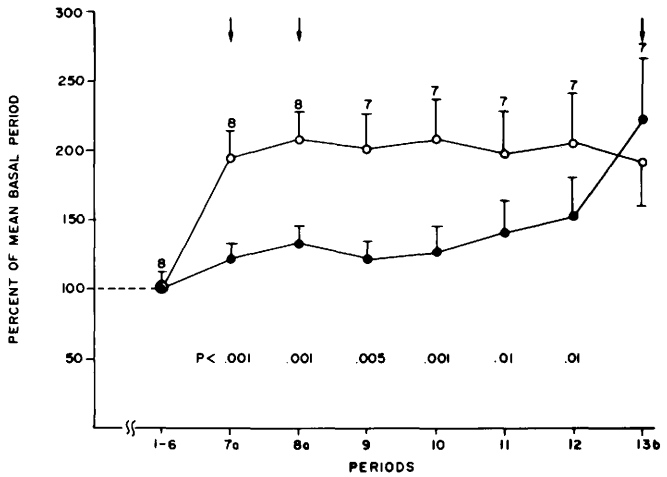

FIG. 1. Antipyrine recovery during treatment periods (7–12), expressed as a percentage of the mean basal antipyrine recovery for periods 1–6, which is assigned a value of 100% and is shown by the dashed line. Test side indicated by open circles; control side by closed circles. The arrows indicate detergent application to test (periods 7 and 8) and control (period 13) hemichambers. Standard error of the mean is indicated by vertical lines. The number of experiments and *P* values are given.

pressed as a percentage of basal drug recovery. Following detergent treatment, antipyrine recovery significantly increased on the test side when compared with the control. Although detergent was placed on the test side only during periods 7–8, the effect persisted throughout the experiment. After the twelfth period the control side was treated with detergent. This produced a substantial rise of antipyrine recovery which exceeded that of the test side.

The effect of detergent treatment on the gastric clearance of aminopyrine, expressed as a percentage of basal drug recovery, is presented in Fig. 2. Following detergent treatment, aminopyrine clearance significantly increased on the test side compared with the control. The differences tended to diminish in the later periods of the experiments.

The effect of detergent on gastric mucosal permeability is presented in Fig. 3. Following applications of detergent to the test hemichamber a fall in the gastric mucosal potential difference, loss of instilled hydrogen ions and increase in sodium secretion was seen in the treated side as compared to control. Only trace amounts of hemoglobin were detected in the test solutions (0.025 mg/per sample).

Discussion. The major determinants of transport of drugs from the systemic circulation to gastric juice are the pK_a and protein-binding of the drug, lipid solubility, gastric blood flow, and permeability of the epithelial surface (1, 2).

It has been well established that biologic membranes are more permeable to uncharged molecules (unionized) than to those bearing an electrical charge (ionized) (15). The pK_a of an acid or base is defined as the pH of an aqueous solution at which the substance is half ionized and half unionized. Acids exist largely in the unionized state and bases in the ionized states at pH's below the pK_a, whereas the reverse is true at pH's above the pK_a. Therefore, drugs which are unionized at plasma pH readily diffuse across the gastric mucosal cells which behave as a lipoidal barrier. Having entered gastric juice, which has a lower pH than plasma, the drugs are ionized, less lipid soluble and are thereby prevented from diffusing back across the gastric mucosa. This phenomenon is referred to as ion-trapping and is intergral to the pH-partition hypothesis derived as an explanation for the unequal distribution of substances between gastric juice and plasma. Accordingly, a number of basic organic compounds, particularly with a pK_a between 4.6 and 5, have been found in considerably greater concentrations in

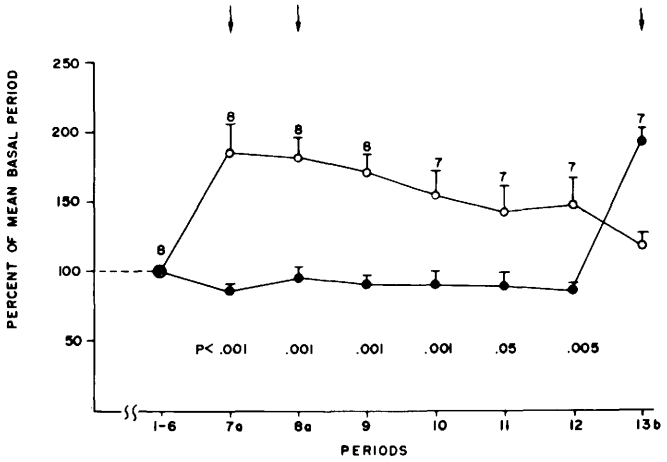

FIG. 2. Aminopyrine clearance during treatment periods (7–12), expressed as a percentage of the mean basal clearance for periods 1–6, which is assigned a value of 100% and shown by the dashed lines. Test side indicated by open circles; control side by closed circles. The arrows indicate detergent application to test (periods 7 and 8) and control (period 13) hemichambers. Standard error of the mean is indicated by vertical lines. The number of experiments and *P* values are given.

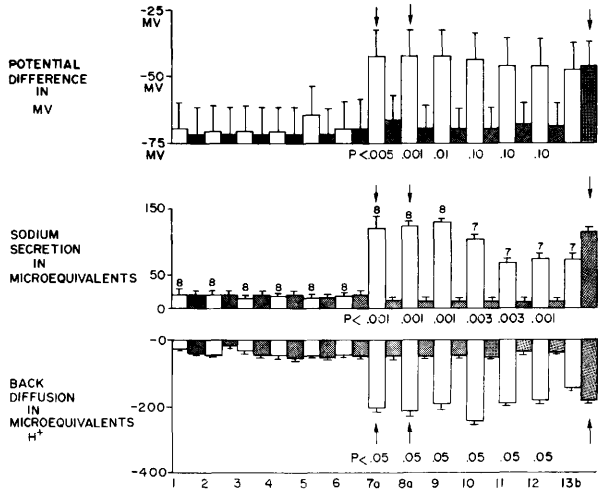

FIG. 3. Gastric mucosal potential difference, sodium secretion, and hydrogen ion back-diffusion during basal and treatment periods. Test side indicated by open squares; control side by shaded squares. The arrows indicate detergent applications to test (periods 7 and 8) and control (period 13) hemichambers. Standard error of the mean is indicated by vertical lines. The potential difference and hydrogen ion back diffusion was studied in three experiments. Sodium secretion was studied in the number of experiments shown. The significance of differences between control and treated hemichambers following detergent treatment are given.

gastric juice than in plasma. The limiting factor of drug transport in this situation appears to be gastric blood flow and protein binding.

A number of studies have shown that detergents alter the permeability of biological membranes and reduce the resistance of a lipid bilayer membrane. Zutphen *et al.* (6) showed that sublytic concentrations of polyoxyethylene ether detergents are able to reduce the direct current resistance of lipid bilayer membranes and enhance their permeability to cations. Moore *et al.* (7) demonstrated that surfactants increased in-

testinal permeability to glucose *in vitro*. Anello and Levy (5) found that low concentrations of the nonionic surfactant polysorbate 80 significantly increased the absorption and exsorption rate constants of four amino-antipyrine in goldfish. Ostrow (15) found that bile salt-induced gall bladder mucosal injury facilitated absorption of ionized molecules. In man, Myerson *et al.* (16) showed a 50–80% increase in riboflavin absorption when 600 mg of sodium deoxycholate was concomitantly administered. Accordingly, we treated dog gastric mucosa with the detergent sodium lauryl sulfate in order to assess its effects on the transport of the drugs antipyrine and aminopyrine.

Under the conditions of our study we found that treatment of the gastric mucosa with detergent significantly increased transport of antipyrine and aminopyrine across the gastric epithelium. Since only small amounts of hemoglobin were present in the solution recovered from the detergent treated hemichamber, the increase in antipyrine transport cannot be ascribed to hemorrhage from an injured mucosa. Two explanations for the increase of drug transport seem worthy of consideration: (i) an increase of gastric mucosal permeability and/or (ii) an increase in gastric mucosal blood flow. Disruption of the gastric mucosal barrier was evidenced in our experiments (following detergent treatment) by the increase in sodium recovered from the instilled solutions, increased back diffusion of hydrogen ions, and a decrease in the gastric mucosal potential difference. This is consistent with the findings of Davenport and others (17–20), who have demonstrated an increase in back diffusion of hydrogen ions associated with increased sodium secretion following treatment of the gastric mucosa with natural and synthetic detergents. The disrupted gastric barrier is commonly associated with epithelial cell damage (20, 21) and, according to Auger (22), an increase of aminopyrine clearance which has been considered a measure of gastric mucosal blood flow (23, 24). Aminopyrine clearance increased significantly in our experiments following application of detergent to the gastric mucosa. Since antipyrine is a nondissociated lipid soluble molecule at plasma pH and is not bound significantly to plasma proteins, an increase in gastric mucosal blood flow may be the major determinant of the increase of drug transport in our experiments.

In order to define further the precise role of gastric blood flow and alterations in mucosal permeability, we are currently studying the effect of detergent treatment of the gastric mucosa on transport of drugs which are ionized at plasma pH and on transport under conditions of maximal gastric secretion and maximal gastric mucosal blood flow.

Summary. The effect of detergent treatment of the canine gastric mucosa on transport of drugs from blood to gastric juice was studied using a chamber technique, *in vivo*. Detergent treatment was found to increase antipyrine and aminopyrine transport. Facilitation of drug transport was associated with disruption of the gastric mucosal barrier and an increase of aminopyrine clearance.

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